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Net Exchange of Certain Inorganic Ions and Water in the Alimentary Tract of the Milk-Fed Calf

By R. H. SMITH

National Institute for Research in Dairying, University of Reading

(Received 6 October 1961)

The present work was undertaken initially to study the effective sites of magnesium and calcium absorption in the alimentary tract of the milk-fed calf and to examine further the decrease previously observed (Smith, 1957, 1958*a*, 1959*c*) in the net absorption of these substances with increasing age.

Magnesium is of particular importance in cattle (and sheep) nutrition since, except in very young calves (Smith, 1957, 1958*a*, 1959*c*), it appears to be poorly absorbed by these animals from normal diets (Blaxter & McGill, 1956; Rook & Balch, 1958; Field, McCallum & Butler, 1958), and clinical deficiency sometimes occurs under normal management conditions. The little that is known of the mechanism of magnesium utilization in the ruminant, mainly from experiments with sheep, suggests that the small intestine is the principal site of absorption (Stewart & Moodie, 1956; Field, 1961). This was also found by Graham, Caesar & Burgen (1960) for the human. The rumen does not appear to be an important site when the rumen fluid contains magnesium in concentrations within normal limits (Storry, 1960; Smith, 1959*a*).

Knowledge of the sites of calcium absorption is in a similarly fragmentary state. The rumen again appears to be unimportant under normal conditions (Storry, 1960), and absorption in the small intestine, found for rats (e.g. Nicolaysen, 1951), has also been demonstrated in the sheep (Storry, 1960),

particularly from the duodenal end. However, several workers have reported calcium absorption in different animals from calcium-containing suppositories inserted in the large intestine (Braun, 1948; Marolda, 1954). Evidence from rat experiments suggests that the small intestine is an important site for the action of vitamin D (Nicolaysen, 1951), but Harrison & Harrison (1951) reported that it had its greatest effect in the more distal parts of the intestine where calcium solubility was low, and Marolda (1954) reported that calcium absorption from the human rectum was increased on vitamin D administration.

I have used suitably fistulated calves of different ages fed basically on milk with, in some experiments, supplementary vitamin D and, in others, different dietary levels of magnesium, to examine the amounts of magnesium and calcium appearing at different points in the alimentary tract relative to an inert marker (polyethylene glycol). A similar examination has been carried out for potassium, sodium, chloride, water and, in a more limited way, inorganic phosphate. This was done both for its intrinsic interest and in the hope of finding changes related to the changes which occur in magnesium absorption with age. Suggestions that other ions affect magnesium absorption, based either on direct experiment or interpretations of secondary effects, have been made for potassium (Kunkel, Burns & Camp, 1953), sodium (Ross, 1961),

phosphate (O'Dell, Morris, Pickett & Hogan, 1957) and calcium (Alcock & MacIntyre, 1960).

A brief preliminary account of part of this work has been published (Smith, 1959b).

EXPERIMENTAL

Animals

Except for calves 9C and 10C which were Hereford $\times$ Shorthorn crossbreds, all the calves used in the present study were Friesians. All were uncastrated bull calves. They were removed from their dams at about 4 days of age and then bucket-fed with 4.40 l. of milk/day. Milk intake was increased to 6.15 l./day from 10 weeks of age. A mineral supplement containing iron, copper and manganese, as described by Smith (1957), was added from 3 weeks of age. Polyethylene glycol (mol.wt. 4000) and vitamin D, in amounts described below, were also sometimes added. The calves were efficiently muzzled (Smith, 1961a) from about 1 week of age. Each day milk samples (10 ml.) were added to 10% (w/v) trichloroacetic acid (40 ml.); the mixture was kept for 2 days at 39°, filtered and the filtrate stored for subsequent analysis. One of the calves was given low-magnesium synthetic milk for part of the time. This milk was prepared essentially as described by Smith (1959c) except for the following modifications. The casein was provided as calcium caseinate (Casilan, Glaxo Laboratories Ltd.) and 2.27 kg. of this was added slowly with continuous mechanical stirring to about 30 l. of water containing 26.4 g. of NaOH and 24.0 g. of citric acid. Stirring was continued until solution was complete (about 2 hr.). Glucose, emulsified margarine, vitamins (except that vitamin D was omitted and added separately to each feed in the amounts described below) and the trace elements iron, manganese, copper and cobalt were then added as described previously and the volume of the mixture was made up to 68 l. This was stored as before and 3.08 l. amounts taken as required for feeding. To each individual feed was added a mineral mixture prepared as follows. Three solutions were made: solution A, 22 g. of NaOH and 20 g. of citric acid in 1 l.; solution B, 11 g. of Na_2HPO_4 , 70 g. of K_2HPO_4 and 15 g. of KH_2PO_4 in 1 l.; solution C, 142 g. of $\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$ in 1 l. For addition to a feed of 3.08 l. of synthetic milk, 84 ml. of solution A, 140 ml. of solution B and 70 ml. of solution C were mixed together. A bulky precipitate of calcium phosphates appeared (probably mainly hydroxyapatite) but this readily dispersed as a fine suspension when the mixture was added to the feed. Thus a uniform calcium (and phosphate) intake was assured from feed to feed. When, as in previous work, the major minerals were added to the synthetic milk bulk, a calcium-containing precipitate was formed which could not be uniformly sampled from feed to feed. The present procedure gave a net composition of the final mixture close to that of whole milk with respect to the major minerals (except magnesium) and the pH was about 7.0.

Many of the calves were fistulated in the small intestine. A cannula, which consisted of an externally threaded Perspex tube about 7 cm. long and 1 cm. internal diam., was secured inside by screwing into a central hole in a Perspex bar (7 cm. $\times$ 1.5 cm. $\times$ 0.4 cm.) which was inserted along the interior of the intestine. The cannula was secured outside with a Perspex nut over a polythene washer (about

10 cm. diam.). Two types of fistula, duodenal and ileal, were employed.

For a duodenal fistula, a single cannula was inserted just below the junction of the abomasum and duodenum and above the bile duct. The cannula was closed with a rubber stopper and samples were obtained by simply removing this stopper. The operation was carried out on calves 1B, 3B, 5B, 14B and 15B at 1–2 weeks of age.

Preliminary experiments showed the impracticability of obtaining samples from the distal end of the small intestine with the type of cannulation used in the duodenum. Only a few ml. were obtained after collection over several hours so that it was impossible to relate a sample to a particular collection time. The following procedure for an ileal fistula was therefore adopted. The intestine was severed just above the junction of the ileum and caecum and a cannula was inserted into each of the severed ends. The cannulae were joined outside the animal by a flexible tube. Collections of the total flow from the small intestine were then made by disconnecting this tube and running another tube from the efferent cannula into a collecting vessel. In this way, with the calf suitably restrained, complete collections for up to 24 hr. were made. The cannulae were inserted into calves 9C, 10C, 4D and 8D at 1–2 weeks of age and into calves 2D, 3D and 7D at 9–10 weeks of age.

None of the calves used in these experiments showed any apparent impairment of health after the operation and they all grew and behaved normally. The life of a cannula varied considerably but each eventually came out or leaked so badly as to be useless, and the calf was then slaughtered.

Analyses

Polyethylene glycol. Samples from the small intestine were homogenized in an Ato-Mix blender (Measuring and Scientific Equipment Ltd.) and faeces samples were similarly homogenized after adding 3 vol. of water. Samples of the homogenates were centrifuged until the supernatant liquid was reasonably clear. Determination of polyethylene glycol in the faeces supernatant liquid was according to Smith (1959a), and in the supernatant liquid of the small-intestine samples essentially according to Smith (1958b). The latter procedure was, however, modified in later determinations to eliminate the necessity of checking pH on the addition of mercuric chloride. Just before this addition 0.1 ml. of 0.04% thymolphthalein was added to the mixture in a 10 ml. volumetric flask and mercuric chloride solution was added until the deep-blue colour was discharged. The mixture was made up to volume, filtered and treated as before.

Calcium and magnesium. A sample (about 5 ml.) of an homogenate of small-intestine contents or faeces was added to a weighed silica crucible by means of a wide-tipped pipette. The crucible was weighed, dried on a steam bath and in an oven at 98° overnight and re-weighed. The dried residue was digested with 2 ml. of concentrated HNO_3 for 1 hr., dried and then ashed at 500° overnight. Occasionally a further treatment with concentrated HNO_3 and further ashing were required. The ash was taken up with 2 ml. of 2N- HNO_3 , and the mixture was made up to a suitable volume and subsequently treated as described for faeces by Smith (1957). Calcium and magnesium in milk were determined as described by Smith (1957).

Sodium and potassium. A sample of the supernatant fluid obtained by centrifuging homogenates of small-

intestine contents or faeces was suitably diluted, filtered and used directly for sodium and potassium determination with an EEL flame photometer. Preliminary experiments showed that percentage recoveries of sodium and potassium added to such samples (three experiments) were $98, \text{s.d.} \pm 1$, and $100, \text{s.d.} \pm 1$, respectively. Ashing the supernatant fluid (at 450°) had no effect on the potassium determination and increased the observed sodium value by 3, $\text{s.d.} \pm 1$ (mean of three experiments). Determination of sodium and potassium in milk was carried out in the same way after diluting a sample of the trichloroacetic acid filtrate. Percentage recoveries of added sodium and potassium (six experiments) were $102, \text{s.d.} \pm 1$, and $100, \text{s.d.} \pm 0$, respectively.

Chloride. This was determined in samples of the supernatant fluid obtained by centrifuging homogenates of small-intestine contents or faeces and in milk-trichloroacetic acid filtrates by the method of Sanderson (1952). The percentage recovery of chloride added to samples of small intestine contents (11 experiments) was $99, \text{s.d.} \pm 0.5$.

Inorganic phosphate. Determination of phosphate in milk-trichloroacetic acid filtrates was carried out by the method of Fiske & Subbarow (1925). Direct determination of phosphate in small-intestine contents could not be carried out similarly because the commonly used deproteinizing procedures did not efficiently remove interfering materials. The following procedure was used for samples from the proximal end of the small intestine (it was not suitable for samples from the distal end). A 2 ml. sample of the homogenized material was diluted to 10 ml., and 1 ml. of the diluted homogenate was added to a 15 ml. conical centrifuge tube. To this were added 4 ml. of 0.3N-Ba(OH)_2 and 3 ml. of $10\% \text{ (w/v) BaCl}_2$. The mixture was well shaken and allowed to stand for 24 hr., after which it was centrifuged. The supernatant liquid was poured off and the residue washed once with 1 ml. of 0.3N-Ba(OH)_2 and 1 ml. of $10\% \text{ BaCl}_2$ added together. The washed residue was taken up by adding 4 ml. of a solution containing $20\% \text{ (w/v) trichloroacetic acid}$ and $5\% \text{ (w/v) Na}_2\text{SO}_4$; the mixture was made up to 10 ml., well shaken and filtered. Phosphate was determined in the filtrate by the method of Fiske & Subbarow (1925). The percentage recovery of inorganic phosphate (KH_2PO_4) added to samples of contents from the proximal end of the small intestine (three experiments) was $98, \text{s.d.} \pm 2$.

Expression of results. In view of the heterogeneous nature of the small-intestine contents and faeces and the fact that some of the constituents examined were sometimes present to an unknown degree in an insoluble form, all results were calculated in relation to the water present. When a known amount of the total homogenate was taken for analysis (calcium, magnesium and phosphate determinations), the amount of water present was assumed to be given by the loss of weight on drying at 98° . It is unlikely that appreciable amounts of other volatile constituents were present in the samples examined. When a known volume of supernatant fluid was taken for analysis (polyethylene glycol, potassium, sodium and chloride determinations), the determined concentrations in relation to the fluid volume were multiplied by an empirical factor of 1.025 for small-intestine contents and 1.035 for faeces. These factors were mean values (six experiments in each case) found to relate supernatant fluid volumes and their weight losses on drying at 98° .

RESULTS

Polyethylene glycol as a reference substance in the alimentary tract

Earlier experiments have shown a fairly good recovery of polyethylene glycol after passage through the calf alimentary tract (Smith, 1959a). Further experiments have been carried out in a similar way by giving milk-fed calves (not fistulated) single doses of polyethylene glycol ($5\text{--}7 \text{ g.}$) and collecting faeces until two successive samples were virtually free of polyethylene glycol. (Collection for about 8 days was usually required.) Considering these and the previous experiments (in all, 11 experiments with seven calves aged 0.5–16 weeks), the percentage of polyethylene glycol recovered varied from 77 to 103 with a mean value of $89, \text{s.d.} \pm 2.5$. There were no apparent differences in recovery at different ages. The mean percentage recovery for four experiments with four calves aged 0.5–4 weeks was $88, \text{s.d.} \pm 4.5$, and for six experiments with four calves aged 10–16 weeks it was $88, \text{s.d.} \pm 4.0$. The values are rather lower than those for adult bovines (mean 94) found by Hydén (1956).

It is clear that no accurate figures can be obtained for the time of passage through the whole alimentary tract. However, as with most milk-fed calves, six of the seven examined here defaecated every 1 or 2 days. Mean values for polyethylene glycol excreted, expressed as percentages of the total polyethylene glycol excretion, were $46, \text{s.d.} \pm 8$, after 2 days and $95, \text{s.d.} \pm 2$, after 4 days in nine experiments with these calves. Occasionally calves void faeces much less frequently, in extreme cases at intervals of 8–10 days. The remaining calf examined here behaved in this way, and polyethylene glycol appearance was much delayed.

Experiments to determine recovery of polyethylene glycol after passage through the alimentary tract up to the distal end of the small intestine were carried out with calves equipped with re-entrant cannulae at this point and regularly fed on milk containing a supplement of polyethylene glycol ($1.14 \text{ g./l. of milk}$). A complete collection from the disconnected efferent cannula was begun at the time of a morning feed (about 9 a.m.) and continued for 24 hr., the calf being fed again at about 5 p.m. Fig. 4 shows a typical pattern of emergence of polyethylene glycol during the first 7 hr. after the morning feed. A similar pattern was observed in all experiments, with calves aged from 2–23 weeks, in which milk supplemented only with polyethylene glycol, and in some cases vitamin D also, was given. Table 1 shows the estimated times after feeding at which dietary polyethylene glycol first appeared in the effluent and

the percentage of dietary polyethylene glycol appearing in the first 7 hr. after feeding. As long as the flow from the ileum to the colon had previously been unrestricted, samples obtained during the first hour after feeding always contained less than 1% of that given in a feed. This suggested virtually complete elimination in the 17 hr. between the evening feed on one day and the morning feed on the next. Thus the polyethylene glycol surviving from the two feeds given on any one day should be accurately represented by that collected during the appropriate 24 hr. period. In 17 experiments with six calves (9C, 10C, 2D, 3D, 4D and 7D, aged 2–19 weeks) examined when they were showing no obvious signs of leakage from around the cannulae, the mean percentage recovery was 97.0 , $\text{s.d.} \pm 1.5$. In fact only one calf (10C) out of the six gave recoveries at any time less than 95%. It seems probable that in this calf, which gave recoveries of only 83 and 85% in two experiments, either external leakage occurred unnoticed or an internal connexion between the ileum and colon was re-established (this occurred and was detected in another calf). Omitting calf 10C, the mean percentage recovery was 98.5 , $\text{s.d.} \pm 0.5$. It appears, therefore, at least with the majority of calves, that very little polyethylene glycol is lost up to the end of the small intestine and that this substance is a very satisfactory reference substance in studies up to this point. The poorer recovery in the faeces, by amounts which in many cases were too great to be explained by the experimental difficulty of quantitatively recovering small amounts of faeces, must be borne in mind in interpreting results involving this region. The reason for the apparent frequent loss of polyethylene glycol in the large intestine is not clear. No loss occurred in up to 11 days when polyethylene glycol was incubated at 39° *in vitro* both aerobically and anaerobically with samples of faeces from three different calves. No polyethylene glycol was detected in the urine after calves were given milk containing this substance (Smith, 1959a), but small amounts might have gone undetected in the large volumes of urine involved.

It should be pointed out that all the present observations refer to milk-fed calves. In such calves, irrespective of age, most of the diet usually bypasses the rumen and goes directly to the abomasum (Smith, 1959a). Rumen involvement would probably not affect polyethylene glycol recovery (Sperber, Hyden & Ekman, 1953), but it would almost certainly affect transit time.

Contents entering the duodenum from the abomasum

Calves at different ages, with fistulae in the proximal duodenum, were examined after receiving milk supplemented only with polyethylene glycol. Samples, referred to subsequently as abomasal effluent, were obtained from the cannulae at different times after feeding.

pH values. In 18 experiments with five calves aged 2–20 weeks, mean pH values were 4.23 , $\text{s.d.} \pm 0.10$, and 2.31 , $\text{s.d.} \pm 0.06$, for abomasal effluent samples taken 1 and 6 hr. respectively after feeding. By 6 hr. after feeding it appeared that a minimum value had been reached. The above values were shown apparently irrespective of age, and although the rate of fall of pH varied in different experiments (see Fig. 1) these differences did not appear to be related to age. They may, as is suggested by the results shown in Fig. 1, have been due in part to the ingestion of different volumes of milk relative to the size of the calf. The results given above are all for samples which were white in colour. Calf 15B frequently, and the other calves very occasionally, gave samples which were coloured yellow or green and which showed higher pH values than the white samples. They were obtained most frequently about 6 hr. after feeding, and calf 15B often showed a fall in pH after feeding followed by a rise in the 6 hr. samples to as much as 3.8. It seems highly probable that such samples were contaminated with material regurgitated from below the bile duct. Only results from white samples not showing aberrant pH values are included in the next section.

Minerals and water. Changes in composition with respect to polyethylene glycol in samples of

Table 1. *Emergence of polyethylene glycol in the effluent from the terminal ileum of calves fed on milk containing polyethylene glycol*

The times at which polyethylene glycol first appeared were estimated to the nearest $\frac{1}{4}$ hr. by consideration of curves similar to that illustrated for one calf in Fig. 4. The younger and older calves were given 2.20 and 3.08 l. of milk, containing 1.14 g. of polyethylene glycol/l.,/feed respectively. Two experiments were carried out with each calf within the appropriate age range.

Calves	Age range (weeks)	Time after feeding at which polyethylene glycol first appeared in effluent (hr.)		Dietary polyethylene glycol appearing in the first 7 hr. after feeding (%)		
		Range	Mean	Range	Mean	S.D.
9C, 10D, 4D, 8D	2–5	2–3 $\frac{1}{4}$	3	63–83	76.5	± 2.0
2D, 3D, 7D	11–13	2 $\frac{1}{2}$ –4 $\frac{1}{2}$	3 $\frac{1}{2}$	63–92	75	± 4.5

Fig. 1. Changes, with time after feeding, in the pH of abomasal effluent from a calf (14 B) aged 9–11 weeks. Solid symbols and broken line refer to two experiments in which 0.045 l. of milk/kg. body weight was given. Open symbols and solid line refer to two experiments in which 0.032 l. of milk/kg. body weight was given.

Fig. 2. Changes in abomasal-effluent composition with time after feeding 2.20 l. of milk supplemented with polyethylene glycol to a calf (14 B) aged 2 weeks. Each result is expressed as the ratio of a constituent to polyethylene glycol in the effluent divided by the corresponding ratio in the diet. Ratios to polyethylene glycol in the diet were: water (x), 800; calcium (▲), 1.13; magnesium (△), 0.114; potassium (●), 1.19; sodium (□), 0.59; chloride (■), 1.24; inorganic phosphorus (○), 0.62.

abomasal effluent taken at different times after feeding are illustrated for one calf in Fig. 2. The results for each constituent are expressed as the factor by which the amount of constituent in the effluent differs from the amount in the diet when both are related to a unit amount of polyethylene glycol. Thus values greater than 1 and less than 1 indicate net addition and net removal respectively of the constituent up to the exit from the abomasum. Four other identical experiments with three calves aged 2–10 weeks (5B, 14B and 15B) gave results similar to those shown in Fig. 2. Many more experiments were carried out in which only the samples obtained 1 hr. and, in some cases, 6 hr. after feeding were analysed. These results (mean values in Table 2), for calves aged 2–20 weeks, all showed changes with time similar in direction to those illustrated in Fig. 2. The values for the 1 hr. samples did not show much variation, and, as the results exemplified in Fig. 2 suggested that little change in composition occurred during the first hour, it appeared that an initial net dilution of the milk entering the abomasum [probably usually at least 90 % of that fed (Smith, 1961a)] with 0.3–0.5 part of endogenous fluid occurred fairly uniformly. Values for the 6 hr. samples, varied considerably. The largest mineral variations, which were for potassium, sodium and chloride, were closely related to differences in the degree of dilution of polyethylene glycol and were apparently random variations not associated to an appreciable extent with calf age or calf-to-calf differences. They could be explained by considering the residue of the dietary intake left in the

Table 2. Ratios of various constituents to polyethylene glycol in samples of abomasal effluent divided by the corresponding ratios in the diet

The mean values ($\pm$ S.D.) given were derived from 25 experiments with calves 1 B, 5 B, 14 B and 15 B carried out at ages ranging from 2 to 20 weeks. Effluent samples were taken at different times after feeding milk (2.20–3.08 l.) containing supplementary polyethylene glycol. Mean ratios of the constituents to polyethylene glycol in the diet with standard deviations where appropriate were: water, 800; calcium, 1.09 ± 0.02 ; magnesium, 0.104 ± 0.002 ; potassium, 1.35 ± 0.02 ; sodium, 0.53 ± 0.02 ; chloride, 1.18 ± 0.04 ; inorganic phosphorus, 0.61 ± 0.01 .

	Abomasal effluent 1 hr. after feeding (22 samples)	Abomasal effluent 6 hr. after feeding (17 samples)
Water	1.44 ± 0.04	3.70 ± 0.44
Calcium	0.71 ± 0.02	1.80 ± 0.16
Magnesium	0.91 ± 0.02	1.47 ± 0.12
Potassium	1.04 ± 0.02	1.58 ± 0.23
Sodium	1.88 ± 0.09	6.75 ± 1.16
Chloride	2.44 ± 0.14	11.1 ± 1.6
Inorganic phosphorus	0.90 ± 0.03	2.18 ± 0.25

abomasum 6 hr. after feeding to have been diluted with differing amounts of a fluid essentially constant in composition with respect to potassium, sodium and chloride. This was true although, as is discussed below, the processes which established this net secretion must have been very complex and the total net secretion varied in different experiments by as much as eightfold. The values for relative net secretion, from which the results given in Table 3 were derived, were calculated as follows.

If the ratio of a constituent to polyethylene glycol in a sample of abomasal effluent divided by the corresponding ratio in the diet is x (e.g. the values given in Table 2 and Fig. 2), then the net secretion of this constituent/unit amount of polyethylene glycol is

$$(x-1) \left(\frac{\text{constituent amount in diet}}{\text{polyethylene glycol amount in diet}} \right)$$

and the relative net secretions of two constituents (net secretion of constituent 1/unit amount of net secretion of constituent 2) is

$$\left(\frac{x_1-1}{x_2-1} \right) \left(\frac{\text{constituent 1 amount in diet}}{\text{constituent 2 amount in diet}} \right).$$

Similar calculations for abomasal effluent samples taken at different times after feeding in individual experiments, exemplified in Fig. 3, showed irregular and usually small changes in the net secretion of sodium relative to that of water; there was an increase in the net chloride secretion relative to that of water (and sodium) during the first few hours after feeding, to a value which subsequently remained fairly constant up to 6-7 hr. These findings are supported by a comparison of the mean values given in Table 3 for the 1 hr. and 6 hr. abomasal samples respectively. No conclusions could be reached about changes in net potassium secretion with time since values could be estimated with reasonable accuracy only in samples containing a relatively high proportion of endogenous fluid (i.e. in some of the 6 hr. samples).

In abomasal samples taken soon after feeding, calcium, magnesium and inorganic phosphate always appeared in ratios to polyethylene glycol smaller than those in the diet. These ratios increased with time until in samples taken some hours after feeding all were markedly greater than the corresponding ratios in the diet. This effect, which is shown in Fig. 2 and Table 2, is discussed below.

Our results were concerned principally with the relative amounts of the different constituents leaving the abomasum. They did not yield any detailed information about the absolute amounts leaving at different times, as the proportion of the

total flow discharging through a cannula compared with that bypassing the cannula was unknown. However, gross differences were apparent. Just before a morning feed (i.e. after about 16 hr. fasting), and for about 30 min. after this feed, abomasal effluent was discharged from the cannula in very small amounts at long intervals. The amounts discharged increased rapidly after this, and, between 1 and 4 hr. after feeding, 50-100 ml. was usually obtainable in less than 5 min. After this the flow decreased and 6 hr. after feeding it took about 15 min. to collect 50 ml.

Fig. 3. Changes with time after feeding in the net amounts of sodium (solid symbols) and chloride (open symbols) respectively secreted up to the exit from the abomasum/unit net amount of water similarly secreted. Results are given for three experiments in which calves 5B or 14B at ages from 2 to 7 weeks were given 2.20 l. of milk.

Table 3. Net amounts of sodium, chloride and potassium expressed in relation to the net amount of water secreted up to the exit from the abomasum in samples obtained at different times after feeding

Mean values ($\pm$ S.D.) were derived from 14 of the experiments given in Table 2 in each of which abomasal-effluent samples were taken at both 1 hr. and 6 hr. after feeding. These experiments were with calves 5B, 14B and 15B, aged 2-20 weeks. Values are not given for potassium in 1 hr. samples since the amounts involved were too small to give useful results.

	Sodium (mg./100 g. of water)	Potassium (mg./100 g. of water)	Chloride (mg./100 g. of water)
1 hr. after feeding	123 $\pm$ 6	—	480 $\pm$ 21
6 hr. after feeding	130 $\pm$ 6	22 $\pm$ 8	568 $\pm$ 23

Contents leaving the distal end of the small intestine

Calves with a re-entrant cannulation in the distal ileum (9C, 10C, 2D, 3D, 4D, 7D and 8D) were examined at different ages while they were receiving a diet of milk given as two equal feeds per day at 9 a.m. and 5 p.m. Samples, subsequently referred to as small-intestine effluent, were obtained by disconnecting the tube joining the severed parts of the intestine. A uniform supplement of polyethylene glycol (1.14 g./l. of milk) was added to the diet throughout the experiments, starting at least 2 weeks before samples were taken.

pH value. When small-intestine effluent was exposed to the air its pH rapidly increased presumably owing to loss of carbon dioxide. Consequently samples obtained as routine for analysis could not be used for pH determination. Some collections were made by joining a flexible tube to the efferent part of the cannula, filling this with liquid paraffin and immersing the free end in liquid paraffin in a separating funnel. The first 20–30 ml. of effluent obtained was run off and discarded and the subsequent collection used for immediate pH determination. Samples were obtained in the 2–4 and 5–8 hr. intervals after feeding from each of four milk-fed Friesian calves aged 7–9 weeks. There was no significant difference between the two times of sampling and the overall mean (eight samples) was 7.56, s.d. $\pm$ 0.04.

Minerals and water. Variations with time after giving a morning feed in the amounts of different constituents flowing out of the small intestine are shown for one experiment in Fig. 4. Similar experiments (14 experiments with seven calves aged 2–13 weeks) showed marked differences between calves (but not between different experiments on the same calf carried out within 2 weeks of each other) for (a) the time of emergence of

polyethylene glycol (see Table 1) and the constituents associated with it (see below), and (b) the overall amount of constituents, other than polyethylene glycol, discharged (see Table 4). The general pattern was, however, always the same and consistent similarities, exemplified in Fig. 4, were observed as follows. Samples taken within an hour or two of feeding contained little polyethylene glycol and apparently therefore consisted mainly of endogenous material. The sharp increase in the polyethylene glycol emergence rate which occurred some hours after feeding was accompanied by marked increases in the emergence rates of magnesium and calcium and, to a lesser extent, of water. Potassium, sodium and chloride emergence

Fig. 4. Cumulative amounts of different constituents appearing in the small-intestine effluent at different times after feeding milk (3.08 l.) supplemented with polyethylene glycol. The calf (2D) was 11 weeks of age and the milk fed contained (g./l.): water (■), 905; polyethylene glycol (×), 1.14; calcium (△), 1.36; magnesium (□), 0.130; sodium (▲), 0.56; potassium (●), 1.73; chloride (○), 1.31.

Table 4. Ratios of different constituents to polyethylene glycol in 24 hr. samples of small-intestine effluent and in samples of faeces divided respectively by the corresponding ratios in the diet

Mean values ($\pm$ s.d.) are given for each of two age groups. The younger calves (9C, 10C, 4D and 8D, mean body weight 48 kg.) and the older calves (2D, 3D and 7D, mean body weight 75 kg.) received 4.40 and 6.15 l. of milk/day respectively. Mean ratios ($\pm$ s.d. where appropriate) of the constituents to polyethylene glycol in the diet were: water, 800; calcium, 1.13 ± 0.02 ; magnesium, 0.109 ± 0.02 ; potassium, 1.48 ± 0.03 ; sodium, 0.56 ± 0.02 ; chloride, 1.27 ± 0.04 . Two collections of small-intestine effluent were made within 2 weeks for each calf and the mean of the 2 determinations was used to derive the appropriate group mean. The faeces values for potassium, sodium, chloride and water were calculated for each calf from the compositions of a series of faeces samples obtained at about the same time as the small-intestine-effluent samples ($\pm$ 1 week). The manner of estimating faeces values for calcium and magnesium to relate them to the small-intestine-effluent values is described in the text.

	Age range (weeks)	Water	Calcium	Magnesium	Potassium	Sodium	Chloride
Small-intestine effluent	2-5	0.169 $\pm$ 0.025	0.153 $\pm$ 0.026	0.75 $\pm$ 0.07	0.039 $\pm$ 0.008	0.56 $\pm$ 0.10	0.145 $\pm$ 0.013
	11-13	0.210 $\pm$ 0.048	0.231 $\pm$ 0.034	0.70 $\pm$ 0.07	0.066 $\pm$ 0.019	0.66 $\pm$ 0.18	0.138 $\pm$ 0.019
Faeces	2-5	0.0148 $\pm$ 0.0018	0.142 $\pm$ 0.020	0.45 $\pm$ 0.07	0.0063 $\pm$ 0.0010	0.0124 $\pm$ 0.0060	0.0041 $\pm$ 0.0012
	11-13	0.0165 $\pm$ 0.0039	0.268 $\pm$ 0.046	0.61 $\pm$ 0.10	0.0056 $\pm$ 0.0009	0.0080 $\pm$ 0.0027	0.0052 $\pm$ 0.0011

rates remained nearly constant during the whole period, showing little or no relation to the rate of polyethylene glycol emergence.

Thus very marked diurnal variations occurred in the ratios of the different constituents to each other and in particular to the polyethylene glycol used as a marker. These variations were clearly related to the changes in total flow and composition of the material entering the small intestine at its proximal end—changes which, as shown in the preceding section, resulted mainly from the feeding pattern but were modified with respect to calcium and magnesium by the temporary hold-up of these substances in the abomasum. It follows that small samples taken without regard to these diurnal variations would be of little value in assessing overall net absorption in the small intestine.

The appropriate results shown in Table 4 were all based on complete collections (two from each calf) of small-intestine effluent obtained over a period of 24 hr. These mean results are expressed as the ratios of the various constituents to polyethylene glycol in the small-intestine effluent divided by the corresponding ratios in the diet. If it is assumed that polyethylene glycol is a perfect marker then, if this value for any one constituent is y , the net percentage of the intake of this constituent absorbed is $100(1-y)$. Statistically the results showed no significant differences between the younger and the older groups in the net absorptions of the different constituents. There was, however, considerable variation between calves in the same group as shown by the standard deviations given in Table 4. This variation was much greater than that between duplicate determinations on the same calf carried out within 2 weeks. However, in only one of the younger calves (4D) did the cannula survive sufficiently long to make it possible to examine the effluent from the same calf over a wide age interval. For this calf, the net absorptions (calculated as above) of calcium and magnesium were 92 and 28% respectively at 4 weeks and 75 and 22% respectively at 13 weeks. These figures and other considerations which are discussed below suggest that the marked difference between the mean values for calcium in the two age groups shown in Table 4, although not statistically significant ($0.2 > P > 0.1$), indicates a decrease in net calcium absorption with age.

Faecal excretion

With one exception a total of 5–7 faeces samples was obtained from each calf used in the experiments described in the previous section. The samples were taken over a period starting not more than 1 week before the first collection and ending not more than 1 week after the second collection of small-intestine effluent. The exception

was calf 9C for which only two collections of faeces were made, one between and one after the collections of small-intestine effluent. A period of at least 4 days was always allowed between a collection of small-intestine effluent and a subsequent faeces collection since the removal of this effluent would probably temporarily modify the faeces composition.

To calculate the individual results from which the faeces values given in Table 4 were derived, each faeces sample was related to the mean composition of the milk given during the period of 4 days starting 1–3 days before the faeces sample was obtained. This minimized possible errors resulting from day-to-day variations in the concentrations of the different constituents studied in the milk. These variations rarely exceeded $\pm 5\%$ over periods of about 10 days; so, even for calf 8D which defaecated at irregular and sometimes long intervals (2–7 days), the error could not have been very great. The other calves defaecated every one or two days. For sodium, potassium, chloride and water the mean values for the series of faeces samples obtained for each calf were calculated and the results given in Table 4 were derived from those values. However, the individual faeces values for calcium and magnesium showed a consistent tendency to increase with age over the period of examination of any one calf. For these constituents therefore, for each calf, the individual results were plotted graphically (e.g. in Fig. 5) and

Fig. 5. Calcium and magnesium relative to polyethylene glycol appearing in individual successive samples of small-intestine effluent (24 hr. collections) (open symbols) and faeces (solid symbols) from a single calf (10C). The calf was given milk (4.40 l./day) supplemented with polyethylene glycol (5 g./day) throughout the experiment. Results are expressed as the ratio of calcium (Δ , $\blacktriangle$) or magnesium ($\circ$, $\bullet$) to polyethylene glycol in the sample divided by the corresponding ratio in the diet. The arrows indicate the faeces values for this calf used to derive the appropriate mean values in Table 4.

the faeces value shown 2 days (4 days for calf 8D—see above) after the mean time of collecting the two small-intestine-effluent samples estimated. The periods of 2 or 4 days were chosen as giving a reasonable measure of the time of transit in the large intestine. Values obtained in this way were used to derive the calcium and magnesium figures for faeces shown in Table 4.

Calculations of net absorption in the whole alimentary tract from the appropriate values in Table 4 must take into account the loss of polyethylene glycol in the large intestine. If it is assumed that only 89 % is recovered (see above) then the overall net absorption of any one constituent is $100(1 - 0.89z)$, where z is the ratio of that constituent to polyethylene glycol in the faeces divided by the corresponding ratio in the diet. Although there were considerable calf-to-calf variations in the values obtained within the age groups for calcium and magnesium, as shown by the standard deviations given in Table 4, the variations were fairly consistently reflected in both the small-intestine effluent and faeces for each calf in any one group. Thus the estimated net absorption values in the large intestine did not show excessively wide variations within the groups. From the assumptions and calculation indicated above, the net amounts of magnesium and calcium absorbed in the large intestine, expressed as percentages of the intake, were 35 ± 3.5 and 2 ± 1.5 respectively for the younger calves and 15 ± 3.5 and -0.5 ± 1 respectively for the older calves.

Although these values depend upon certain necessary assumptions and approximations as described above, the general picture remains the same whatever reasonable assumptions are made. Similarly, with any such assumptions it is clear

that sodium, potassium, chloride and water were, as a net effect, strongly absorbed in the large intestine of all the calves.

Effect of supplementary vitamin D

It is probable that by the time they were 11–13 weeks of age our calves were deficient in vitamin D; they received winter milk which contains only about 10 i.u. of vitamin D/l. and were kept indoors out of direct sunlight. It has previously been shown that such calves show a marked improvement in overall calcium retention when vitamin D supplements are given (Smith, 1958a). A daily dietary supplement of 70 000 i.u. of vitamin D₃ given to calves 2D, 7D and 4D from 11–13 weeks of age (subsequent to the experiments described in the preceding section) was followed by a marked improvement in net absorption of calcium in the alimentary tract up to the distal end of the small intestine. Table 5 shows that the improvement was rapid for calves 4D and 7D and slower for calf 2D. In most of the calves excessive leakage from around the cannula prevented normal faeces samples being obtained during these experiments, although satisfactory collections of small-intestine effluent could still be made as the leakage mainly occurred from where the afferent part of the re-entrant cannula entered the intestine. However, faeces collection was possible for part of the time with calf 7D. Results for these samples, also shown in Table 5, suggested that calcium exchange in the large intestine was not appreciably affected by the vitamin D supplement.

Net absorption of magnesium did not improve with vitamin D supplementation. In fact there seemed to be a tendency for this to decrease with age, probably, at least in part, because with a

Table 5. *Effect of dietary vitamin D₃ on the amounts of calcium and magnesium relative to polyethylene glycol appearing in samples of small-intestine effluent (24 hr. collections) and faeces*

	Age range (weeks)		Calf 2D Small-intestine effluent	Calf 4D Small-intestine effluent	Calf 7D	
					Small-intestine effluent	Faeces
No supplementary vitamin D	11–13	Calcium	0.273 $\pm$ 0.009	0.253	0.163 $\pm$ 0.013	0.180
		Magnesium	0.75 $\pm$ 0.00	0.78	0.57 $\pm$ 0.04	0.41
Vitamin D ₃ added to diet	14–16	Calcium	0.260 $\pm$ 0.022	0.029	0.074 $\pm$ 0.016	0.071
		Magnesium	0.85 $\pm$ 0.08	0.76	0.68 $\pm$ 0.01	0.48
	19–23	Calcium	0.174	—	0.047 $\pm$ 0.020	—
		Magnesium	0.84	—	0.74 $\pm$ 0.02	—

constant milk intake there was an increase in endogenous magnesium relative to dietary magnesium as the calves grew.

Effect of variations in magnesium intake

In addition to the experiments shown in Table 5 a series of 24 hr. collections of small-intestine effluent was made with calf 7D between 15 and 23 weeks of age while the calf was fed successively with different diets consisting of 6.15 l. of milk supplemented with magnesium chloride or of low-magnesium synthetic milk with and without a supplement of magnesium chloride/day. All diets contained supplementary vitamin D₃ (70 000 i.u./day) and polyethylene glycol (1.14 g./l. of diet). The relationship between the magnesium:polyethylene glycol ratio in the small-intestine effluent and that in the diet is shown in Fig. 6. The results given in Fig. 6 for an intake of unsupplemented cow's milk were based on four experiments, two carried out at the beginning and two at the end of the series of collections. These experiments (represented separately in Table 5) indicated no marked intrinsic change in the calf's ability to absorb magnesium during the period.

An estimate of the magnesium:polyethylene glycol ratio which would obtain in small-intestine effluent with a diet completely free of magnesium was made by back-extrapolating the curve (Fig. 6). From this the value for endogenous magnesium

Fig. 6. Relationship in a single calf (7D) between the magnesium:polyethylene glycol ratio in the diet and that in the small-intestine effluent, with a constant polyethylene glycol intake but varying magnesium intake. The calf received 6.15 l. of diet, containing 1.14 g. of polyethylene glycol/l./day. Results are shown as vertical bars representing the standard deviations about the mean values obtained in replicate determinations. The numbers shown by the results correspond to the following diets: 1, low-magnesium synthetic milk (two collections); 2, cow's milk (four collections); 3, cow's milk + supplementary MgCl_2 (two collections); 4, synthetic milk + supplementary MgCl_2 (two collections).

excretion under these conditions was calculated to be 0.14 g./day or 1.4 mg./day/kg. body weight. Assuming this value to be independent of magnesium intake the true absorption of magnesium, as a percentage of intake, was calculated to be about 48 during feeding with unsupplemented whole milk (magnesium intake 0.7 g./day) and about 30 with whole or synthetic milk supplemented with magnesium chloride (magnesium intake 3.2–3.5 g./day).

Whether whole milk or synthetic milk was given, the calculated values for net calcium absorption up to the distal end of the small intestine were found to be virtually unaffected by supplementing with magnesium chloride. On whole milk, with magnesium intakes of 0.7 and 3.2 g./day, net calcium absorptions, expressed as percentage of intake, were respectively 94, S.D. ± 1 , and 93, S.D. ± 2 ; on synthetic milk, with magnesium intakes of 0.04 and 3.5 g./day, net calcium percentage absorptions were respectively 81, S.D. ± 6 , and 82, S.D. ± 2 . The figures show, however, a marked difference between whole milk and synthetic milk. The reason for this is unknown but it might be relevant that lactose, which is known to influence net calcium absorption (e.g. Vaughan & Filer, 1960), was absent from the synthetic milk.

DISCUSSION

By intermittent sampling along the alimentary tract after giving a feed containing an inert marker, it is possible to obtain information on net exchange of a constituent with respect to dietary intake up to the point sampled, but only at the instants of sampling. This has been done up to the exit from the abomasum. Such information must be related to total flow and integrated to obtain an estimate of net exchange over a protracted period. This has not been done up to the exit from the abomasum but it has been done up to the exit from the small intestine.

Results of the first type are affected if there are differences in the transit rates of the constituents relative to that of the marker. With a milk diet this appears to be so in the abomasum for calcium, magnesium and inorganic phosphorus in relation to polyethylene glycol, the results suggesting that these three substances are temporarily held up at this point. Very probably they are temporarily trapped in the protein-fat coagulum; Verma & Sommer (1957) found that when milk was coagulated with rennin more than half of the calcium and inorganic phosphate and a smaller amount of magnesium were associated with the coagulum. Net exchange across the tract wall up to the exit from the abomasum could not therefore be evaluated for these constituents from the present

results. Net secretion of water, sodium, chloride and, to a small extent, potassium were, however, shown to occur at all times. Two major secretions (saliva and gastric juice) and three compartments (reticulo-rumen, omasum and abomasum, all probable sites for the absorption of one or more of the constituents) could be involved in producing this end effect, but despite this complexity the amounts of endogenous water, sodium and chloride added (as a net effect) to the abomasal effluent remained in remarkably constant proportion to each other, usually with more than twice as many Cl^- as Na^+ ions. This was true for different calves at ages ranging from 2 to 20 weeks, and also for samples taken at different times after feeding except for an increase in the relative amount of chloride after feeding. It seems likely that this increase paralleled the observed decrease in pH and was due mainly to increased secretion of hydrochloric acid into the abomasum. The net amount of chloride relative to that of water added to the abomasal effluent some hours after feeding [mean 570 mg./100 ml. (Table 3)] was similar to the chloride:water ratio in sheep gastric juice given by Masson & Phillipson (1952) (about 570 mg./100 ml.). If it is assumed that a similar amount of chloride occurs in calf gastric juice, an appreciable amount of water must be absorbed up to the exit from the abomasum, since the chloride content of calf saliva is much lower than this [about 125 mg./100 ml. (Kay, 1960)]. Water absorption in this region was also reported by Hydén (1961) and by Masson & Phillipson (1952). These workers supposed the absorption to have occurred in the omasum but the possibility of its occurrence in the abomasum cannot be dismissed. Cope, Blatt & Ball (1943) have shown rapid absorption of deuterium oxide in the stomach of the dog.

The results did not suggest any variation, with increasing age, in the exchange of the constituents examined up to the exit from the abomasum. The pH values found in abomasal effluent were similar to those reported by Mortenson, Espe & Cannon (1935) and did not show any apparent differences due to age. However, all the present calves were efficiently muzzled so that saliva production/unit body weight probably did not increase much during the experiment (Smith, 1961*a*). With calves eating solid food saliva production would increase greatly (Smith, 1961*a*) and this might influence the composition of the abomasal effluent.

The absolute rate at which polyethylene glycol and the other associated food constituents left the abomasum could not be accurately determined from the present results because the total volume leaving the abomasum was unknown. However, rapid flow commenced $\frac{1}{2}$ –1 hr. after feeding and the front of the wave of food constituents can be

regarded as entering the proximal end of the small intestine at about this time. The unabsorbed residue of this front has been shown to reach the distal end of the small intestine $1\frac{1}{2}$ – $3\frac{1}{2}$ hr. later, a time of transit which showed no marked differences due to age and was comparable to that reported by Hydén (1961) for sheep fed on a normal diet (1–2 hr.).

The results show that, with intermittent milk feeding, the composition as well as the amount of material entering the proximal end of the small intestine showed wide diurnal variations. The fundamental difference between the small-intestine absorption of sodium, potassium and chloride on the one hand and of calcium and magnesium on the other was illustrated by the fact that only the latter reflected to an appreciable extent such diurnal variations in their rates of emergence from the distal end of the small intestine. Measurements of unidirectional flux rates have shown that sodium, potassium and chloride all pass readily (although not necessarily by simple diffusion) across the intestinal mucosa in both directions [for references, see Berger (1960)] and it appears probable that these ions were absorbed until an equilibrium existed between the influx and outflux across the intestinal wall irrespective (within my limits) of the composition and flow rate of the abomasal effluent.

The results derived from the constituent:marker ratios in 24 hr. collections of small-intestine effluent and in faeces (Table 4) confirmed preliminary findings (Smith, 1959*b*) that in milk-fed calves net absorption of magnesium up to the exit from the small intestine does not vary greatly with age (at least after about 2–3 weeks), but that net absorption in the large intestine, which is considerable for young calves, decreases markedly with age. The oldest calves examined in the present work (11–13 weeks) apparently still absorbed some magnesium in the large intestine. However, previous balance experiments (Smith, 1961*b*) with intact calves treated similarly to those used in the present work showed an eventual decrease in overall net magnesium absorption (by about 15 weeks) to an apparently constant mean value of about 22 % of intake. This is close to the mean net absorption up to the distal end of the small intestine (about 27 % of intake) estimated for all the calves, young and old, in the present experiments. Consequently it is probable that the large intestine eventually ceases to absorb appreciable amounts of magnesium. No reason for this change was apparent from the present results and no associated variations in absorption of other minerals were observed. Unfortunately, not enough pH determinations were made to give information on possible variations in the solubility of magnesium

salts. Neither was any reason apparent for the great calf-to-calf differences in ability to absorb magnesium up to the end of the small intestine (9–44 % of intake), differences which are similar to those observed for overall absorption (Smith, 1961*b*).

The fact that in a calf (7D) given diets containing different amounts of magnesium the net magnesium absorption up to the end of the small intestine was not quite linear in relation to the intake is not necessarily incompatible with the linear relationship previously reported between intake and overall net magnesium absorption (Smith, 1959*c*). The previous maximum intake (expressed per unit body weight) was only about 60 % of that examined in the present experiment so that deviations from linearity would scarcely have been apparent. Graham *et al.* (1960) have recently found that the absorption of ^{25}Mg in the human small intestine is directly related to magnesium intake but not linearly. Percentage absorption decreased with increasing magnesium intake in a manner similar to the change in estimated true absorption for calf 7D. McAleese, Bell & Forbes (1961) have recently reported that ^{25}Mg was absorbed more efficiently by lambs on a magnesium-deficient diet than by control lambs. The earlier finding from balance experiments that vitamin D had no appreciable effect on net magnesium absorption by milk-fed calves (Smith, 1958*a*) was confirmed by the present results (Table 5).

Calcium absorption showed quite different characteristics from magnesium absorption. Differences between calves in overall calcium absorption could be attributed almost entirely to corresponding differences in absorption up to the end of the small intestine and it appeared that virtually no calcium was absorbed in the large intestine in either the younger or older calves. The difference between the younger and older calves in mean overall absorption values was, although not statistically significant, presumably a manifestation of the decrease in net absorptive ability with age observed previously (Smith, 1961*b*), a view supported by the values for a single calf (4D) over a wide age range. This was probably due to the depletion of vitamin D stores. The improvement in net calcium absorption up to the end of the small intestine which occurred on adding vitamin D to the diet (dramatic for two calves, less marked for another) did not appear to be accompanied by any change in large-intestine absorption in the only calf for which measurement was possible. It is unlikely that the absence of appreciable net calcium exchange in the large intestine was due merely to a balance of endogenous secretion and absorption since the effect was observed equally

with considerable differences in the amounts of calcium (0.4–2.3 g./day) entering the large intestine. Nevertheless, it is clear that these experiments are not sufficiently precise to exclude the possibility of absorption or secretion of calcium in the large intestine in amounts small compared with the levels employed. The results are not incompatible, therefore, with the fact that large-intestine secretions contain calcium (Storry, 1961) or with absorption from very high local concentrations of soluble calcium salts inserted into the rectum (Braun, 1948; Marolda, 1954). It appears, however, that, under the present conditions, such exchanges are negligible in relation to calcium absorption as a whole.

The above differences in behaviour of calcium and magnesium, together with the fact that, with otherwise similar diets, very great differences in magnesium intake had no appreciable effect on calcium absorption, do not support the idea of a common mechanism for calcium and magnesium absorption suggested by Alcock & MacIntyre (1960) for the rat.

The results for the other constituents studied showed that water, potassium and chloride were, as a net effect, efficiently absorbed in the small intestine although further net absorption of all occurred in the large intestine. The results indicated, however, the importance of the large intestine in sodium absorption. It appeared that the equilibrium conditions at the distal end of the small intestine were such that large amounts of sodium passed into the large intestine where they were efficiently absorbed. This agrees in general with the findings of Hydén (1961) for normally fed sheep.

SUMMARY

1. Calves of different ages were suitably fistulated so that samples could be obtained from different points in the alimentary tract and were fed on milk containing polyethylene glycol as a marker. Preliminary experiments showed this substance to be recovered almost quantitatively from the distal end of the small intestine although a mean loss of about 10 % apparently occurred in the large intestine.

2. In different calves of different ages the net effect of the exchanges of sodium, chloride and water up to the exit from the abomasum was equivalent to the progressive addition of a fluid containing these constituents in approximately constant ratios to each other except that the relative amount of chloride increased somewhat after feeding.

3. Calcium, magnesium and inorganic phosphorus were temporarily held up in the abomasum presumably in association with the milk clot.

4. In young calves (age 2–5 weeks) the mean net absorptions of magnesium up to the distal end of the small intestine and in the large intestine respectively were about 25 and 35 % of the intake. The former figure did not change markedly with age or vitamin D status; the latter decreased greatly with age. It appeared that the true percentage absorption of magnesium up to the end of the small intestine decreased with increasing intake.

5. Mean net absorption of calcium up to the end of the small intestine was about 86 % of the intake in the young calves and apparently decreased with age. Vitamin D markedly improved the absorption in older calves (age about 3 months) but wide variations in magnesium intake had no apparent effect. Net exchange of calcium in the large intestine was negligible at all times.

6. Water, potassium and chloride were, as a net effect, efficiently absorbed up to the end of the small intestine (mean values 80–95 % of intake) but further net absorption of all occurred in the large intestine. Mean net absorption of sodium was only about 40 % of the intake up to the end of the small intestine but almost complete in the large intestine. Net exchanges of these constituents did not vary markedly with age.

7. The pH of the tract contents varied from 4.2 to 2.3 at the exit from the abomasum and was about 7.6 at the distal end of the small intestine.

The author is greatly indebted to Dr A. T. Cowie who carried out all the surgical work required for these experiments. He would also like to thank Mr H. S. Hallett, B.E.M., for assistance with the analytical work and Mrs O. M. Campbell, Miss P. Lewis and Mr B. D. Lawrence for general technical assistance. Thanks are also due to Glaxo Ltd. for a free supply of calcium caseinate.

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