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DIGESTIVE SECRETIONS AND THE FLOW OF DIGESTA ALONG THE DUODENUM OF THE SHEEP

BY F. A. HARRISON* AND K. J. HILL

From the Agricultural Research Council Institute of Animal Physiology, Babraham, Cambridge

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Passage of digesta from the forestomachs of the ruminant is a more or less uninterrupted process and is reflected in the continuous nature of the secretory and mechanical behaviour of the remainder of the tract. Thus secretion of gastric juice, bile and pancreatic juice occurs throughout the twenty-four hours (Hill, 1955; Taylor, 1958; Harrison & Hill, 1960); and the flow of digesta from the abomasum is likewise essentially a continuous process. Quantitative data on the rate of flow of digesta into and from the abomasum are, however, necessary for a correct appraisal of the digestive events which occur in this chamber, and several attempts have been made to obtain this information (Phillipson, 1952; Hogan & Phillipson, 1960; Oyaert & Bouckaert, 1961; Singleton, 1961). The regulation of abomasal secretory activity and its integration with the flow of digesta have also been investigated (Hill, 1960; Ash, 1961*a*), but the activity of the secretory glands associated with the remainder of the tract and their effects on, and interrelationships with, the flow of digesta have so far received little attention. Preliminary investigations on some of these topics in relation to the duodenum are described in this paper.

METHODS

Surgical preparations

One-to-two-year-old Clun Forest sheep were removed from pasture to the animal house where they remained for several weeks before operation. 1000 g chopped hay and 200 g crushed oats were fed once daily at 9.30 a.m. On occasion this ration was divided into three parts: 300 g hay and 100 g oats; 400 g hay; 300 g hay and 100 g oats which were fed at 9.30 a.m., 2.00 p.m., and 5.30 p.m., respectively. At least 1 month was allowed for adaptation to a change in the feeding régime before experiments were done. Mineral supplements were provided for all animals and water *ad libitum*.

Anaesthesia was induced with pentobarbitone sodium ('Nembutal', Abbott Laboratories) and continued with cyclopropane–oxygen in closed circuit (Gregory, 1947).

Rumen fistulae were prepared in all animals and were fitted with polyvinyl cannulae.

Abomasal fistulae and abomasal fundic pouches were prepared as described by Hill (1960).

* Aleen Cust Fellow, Royal College of Veterinary Surgeons, 1959–61.

Abomasal pyloric pouch. The pyloric region was completely transected at two points approximately 5 cm apart and continuity of the tract re-established by anastomosis of the cranial and caudal cut ends of the abomasum. The pouch was prepared by closing each end of the 5 cm segment of pylorus and was fitted with a Perspex cannula inserted through a purse-string suture placed in the mid region of the pouch which was brought out through a stab wound in the abdominal wall.

Intestinal fistulae. Duodenal and jejunal fistulae were prepared in three sheep. For a description of the cannula and the method used to collect mixed bile and pancreatic secretion see Taylor (1960), who developed the method for the collection of pancreatic juice.

Double re-entrant fistulae. In sheep L 104, the duodenum was transected immediately caudal to the pyloric sphincter and a Perspex cannula (external diam. $\frac{3}{4}$ in. (19 mm), internal diam. $\frac{1}{2}$ in. (13 mm)) inserted into each end. The stems of the cannulae were brought out through the body wall and the continuity of the digestive tract re-established by connecting

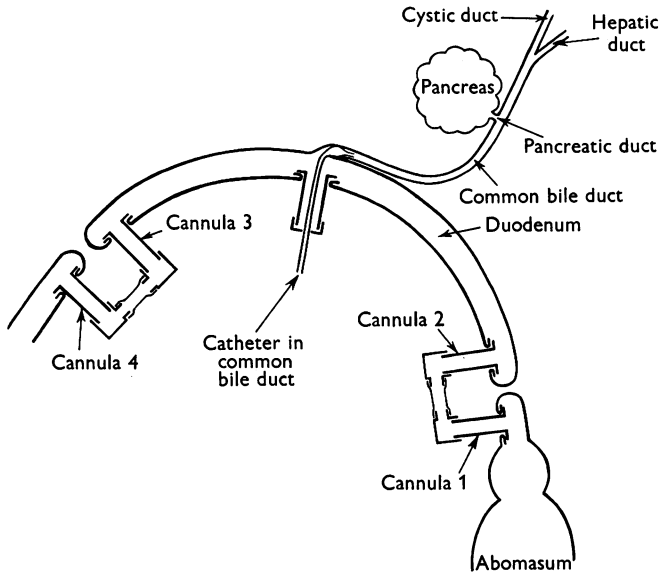

Fig. 1. Diagram of double re-entrant duodenal-fistula preparation.

the two cannulae with plastic tubing. A similar fistula was established about 10 cm caudal to the point of entry of the common bile duct (Fig. 1). It was then possible to isolate a loop of duodenum by disconnecting the tubing between each of the two pairs of cannulae. From this loop of duodenum it was possible to collect and measure bile, pancreatic juice and duodenal secretion. Moreover, by catheterization of the common bile duct it was also possible to collect and measure separately the mixed bile and pancreatic juice and the pure duodenal secretion. Measurement of the rate of flow of digesta through the duodenum was made by collecting the outflow from cannula 1 or cannula 3 in approximately 50 ml. quantities which, after sampling, were returned to cannula 2 or 4 respectively. These experiments were all carried out 3–12 months after the fistulae had been established.

Gall-bladder fistulae were prepared as described by Harrison (1961).

Histological

The duodenum was removed from three normal sheep, immediately after they had been killed with an overdose of 'Nembutal', and fixed in 10% formol-saline. 1 cm blocks

of tissue were embedded in paraffin and sections stained with haematoxylin-eosin and the periodic-acid-Schiff (PAS) technique. Smears of duodenal secretion were stained by Giemsa's stain.

Analytical

pH was determined with a glass electrode (Pye Universal pH meter, Cambridge). Free and total acid of the gastric juice was determined by electrometric titration with 0·1 N-NaOH to pH 3·5 and 9·6, respectively. Peptic activity was estimated by the method of Hunt (1948).

RESULTS

Histological observations on the duodenum

Brunner's glands were found from the pyloric sphincter to the region of the papilla of Vater. In the first few centimetres of duodenum the glands occupied most of the thickness of the mucosa and were present both on the epithelial and serosal side of the muscularis mucosae, which in this region had a disjointed appearance. The ducts of Brunner's glands opened directly on to the epithelial surface and were particularly numerous in this region. A few centimetres along from the pyloric sphincter Brunner's glands occupied a more typical position beneath the muscularis mucosae and in transverse sections of the duodenum appeared as a continuous ring of glands with many ducts which opened on the epithelial surface. Further along the duodenum the glands gradually became separated into discrete collections of alveoli, each with several ducts. The general arrangement appeared to be that of a solid mass of glands adjacent to the pyloric sphincter which extended caudally as a series of tapering fingers of glandular tissue.

The glands were formed of alveoli lined with cuboidal or columnar epithelium which rested on a delicate reticular basement membrane. A few pink-stained cells which resembled parietal cells were seen in the epithelium and globule-leucocytes (Keasbey, 1923) were also present between the columnar epithelial cells. Fibroblasts, plasma cells and lymphocytes were present in the interglandular connective tissue whilst patches of lymphoid tissue were relatively common throughout the duodenal mucosa. The columnar cells lining the ducts contained PAS-positive granules but these were fewer in the cuboidal cells deeper in the ducts and in the actual glandular cells.

Duodenal secretion

Duodenal secretion was collected from sheep L104 after washing the duodenal loop free of digesta with warm physiological saline (NaCl 0·9 g/100 ml.). As the loop contained nearly all the Brunner's glands, the secretion collected represented almost the total output of these glands plus secretion from the epithelial mucous cells. Microscopical examination of smears made from the duodenal secretion and from the deposit, after centrifuga-

tion of this secretion, revealed large numbers of epithelial cells and bacteria. A few eosinophil leucocytes and globule leucocytes were also present. During experiments, the digesta which flowed from cannula 1 were collected and their volume recorded every hour; they were not returned to the duodenum.

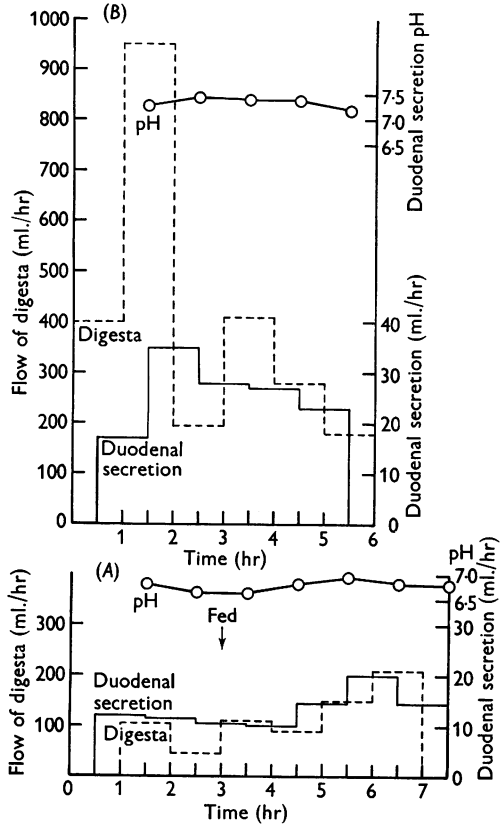

Fig. 2. Flow and pH of duodenal secretion and flow of digesta from cannula 1 of sheep L 104. (*A*) Once-daily feeding; (*B*) thrice-daily feeding.

The secretion collected from the loop was usually colourless in appearance and contained strands of mucus and cellular debris. In one experiment (No. 249), carried out when the animal was conditioned to once-daily feeding, duodenal secretion was collected for 3 hr, food was given and collection continued for an additional 4 hr. The average hourly secretion over the whole experiment was 13·3 ml. and there was a slight increase in secretion after feeding. The average pH after exposure to air was 6·7. The flow of digesta from cannula 1 was very low and averaged 100 ml./hr (range 0–210 ml./hr; Fig. 2*A*). In another experiment (No. 272), when the animal was conditioned to receiving food three times daily, the average

hourly secretion from the duodenum during the 5 hr of the experiment was 26·0 ml. and the pH was 7·35. The animal was not fed during this experiment but the volume of digesta collected from cannula 1 was approximately four times greater than in experiment 249. An exceptionally large volume of digesta was collected during the second hour of experiment 272 and it was followed in the subsequent hour by what appeared to be inhibition of the flow of digesta. The flow of duodenal secretion was greater during the period of increased flow of digesta from cannula 1 and thereafter slowly declined (Fig. 2*B*).

Experiments on the flow of mixed bile and pancreatic juice

In the sheep the pancreatic duct joins the common bile duct, so that a mixture of bile and pancreatic juice flows into the duodenum through the sphincter of Oddi. The combined secretion was collected by temporary catheterization of the common bile duct and the rate of flow determined over 24 hr periods. Figure 3*A* shows the mean hourly volume of secretion collected in four comparable experiments, carried out when the sheep was accustomed to once-daily feeding.

The large volume of secretion obtained in the first hour was probably attributable to drainage of the gall-bladder and the decline in secretion during the second and third hours to interruption of the bile-salt re-circulation which causes a reduction in bile flow (Harrison, 1961). Thereafter the average secretory rate was 38·8 ml./hr or 0·74 ml./kg/hr.

Although these experiments provided information on the flow of mixed bile and pancreatic juice under specified conditions, they gave no indication of the relative amounts of the two secretions present in the mixture. Harrison (1961), however, has shown that bile is secreted at a relatively uniform rate when it is not returned to the intestine and since the mixed secretion was collected under these conditions, comparison of the respective volumes, although they were obtained from different sheep, provided an indication of the relative quantities of bile and pancreatic juice present in the mixture. Thus in eight experiments on four sheep, the average hourly secretion of bile during the 4–24 hr period of the experiments was 0·41 ml./kg/hr compared with an average hourly secretion of mixed juices of 0·74 ml./kg/hr over the same period in four experiments on one sheep. The difference of 0·33 ml./kg/hr can be considered to represent pancreatic juice, and this figure compares with the finding of Taylor (1962) of an average secretion of 0·39 ml./kg/hr of pure pancreatic juice over 24 hr. It should be emphasized that these proportions of approximately equal parts of bile and pancreatic juice are those which obtain during the non-return of secretion to the intestine. As only the rate of bile secretion is depressed by non-return to the intestine (Taylor, 1958; Harrison, 1961) it is evident that under normal

conditions the proportion of bile to pancreatic juice will be greater than that observed here. Thus, in two experiments, when the mixed bile and pancreatic juice were returned to the duodenum, the mean rate of secretion was increased to 53·5 ml. or 0·90 ml./kg/hr during the 4–24 hr period (Fig. 3*B*). Measurements of the total daily flow of each secretion indicate that the proportion of bile to pancreatic juice may be 2 or 3 to 1.

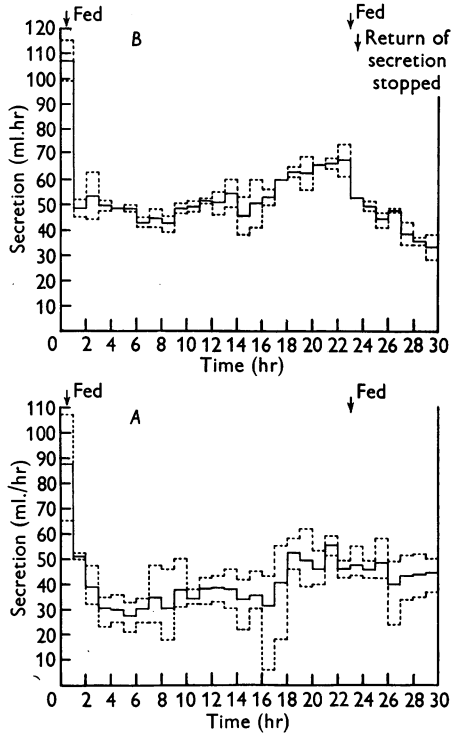

Fig. 3. Mean hourly secretion of mixed bile and pancreatic juice during (*A*) non-return and (*B*) return of secretion to the duodenum.

A change of feeding routine from once daily to one in which the same ration was divided and given on three separate occasions during the day was also associated with an increase in the secretion of bile and pancreatic juice from 25·2 to 34·4 ml./hr over the 4–12 hr collection period. The secretion was not returned to the duodenum in these experiments.

Measurement of flow of digesta from duodenal cannulae

Cannula 1 of sheep L 104 was placed immediately caudal to the pyloric sphincter and hence measurement of the flow of digesta from this cannula provided an indication of the flow from the abomasum to the duodenum.

Hogan & Phillipson (1960) have discussed the errors which may arise when flow rates are measured by direct collection of digesta and their method of minimizing these, in which approximately 50 ml. amounts of digesta are collected and rapidly returned to the duodenum, was adopted in the present experiments.

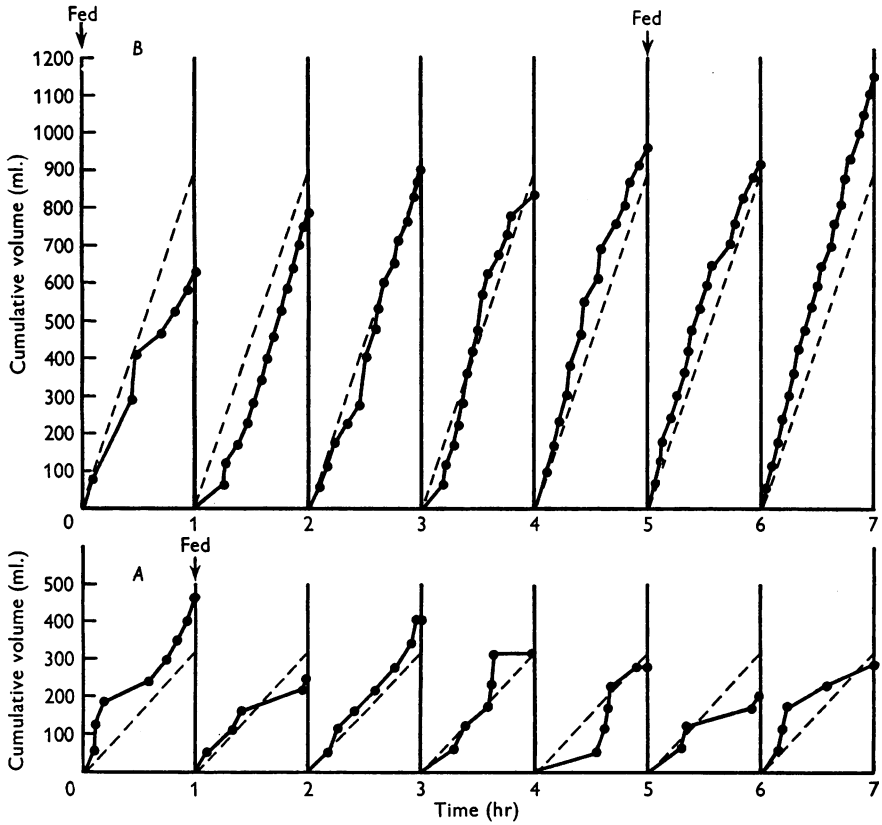

Fig. 4. Cumulative hourly volumes of digesta collected from cannula 1 of duodenal re-entrant fistula during (*A*) once-daily feeding; (*B*) thrice-daily feeding. Sheep L 104.

Measurement of the flow of digesta was first made when the routine feeding procedure was to give the sheep the whole of the daily ration of 1000 g hay and 200 g oats at 9.30–10.00 a.m. each day. It was usual, under these conditions, for the whole of the ration to be consumed in 2–3 hr. During actual experiments the same feeding régime was employed, and in the experiment shown in Fig. 4*A*, the mean flow of digesta from cannula 1, over the 7 hr period of the experiment, was 312 ml./hr. In the figure the time taken for the collection of approximately 50 ml. quantities

of digesta is plotted against the cumulative volume obtained each hour and the interrupted line is the slope of the mean flow of 312 ml./hr.

Further measurements of the flow of digesta from cannula 1 were made after the sheep had become accustomed to a feeding régime in which 300 g hay and 100 g oats were given at 9.30–10.00 a.m., 400 g hay at 2.00–2.30 p.m. and 300 g hay and 100 g oats at 5.30–6.00 p.m. During

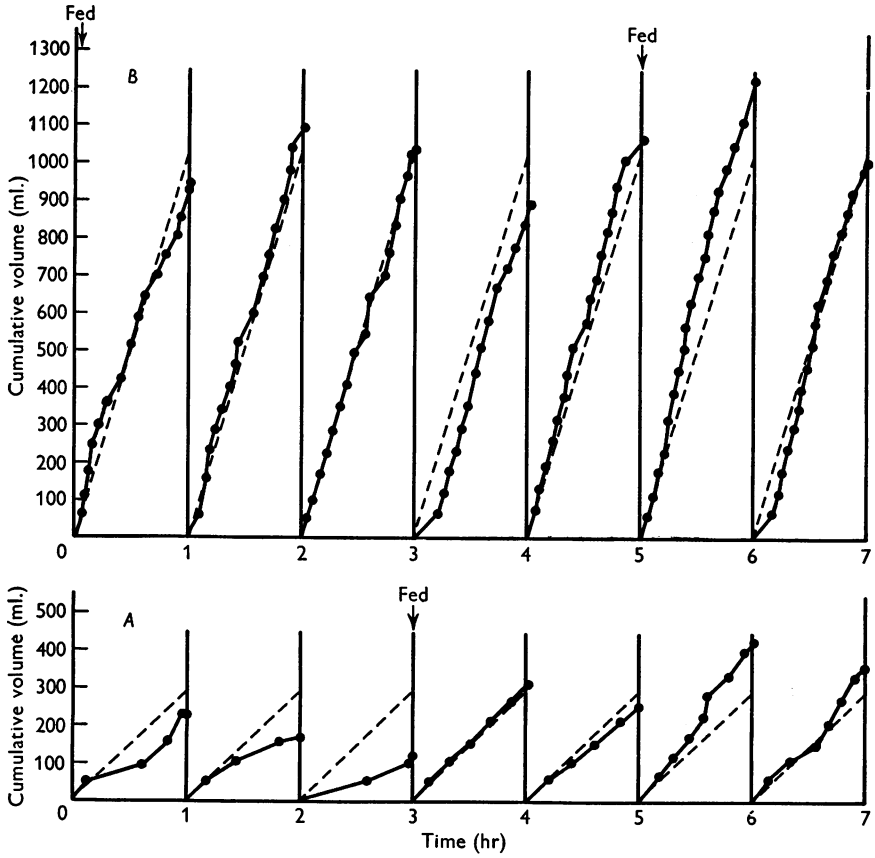

Fig. 5. Cumulative hourly volumes of digesta collected from cannula 3 of duodenal re-entrant fistula during (*A*) once-daily feeding; (*B*) thrice-daily feeding. Sheep L104.

experiments lasting from 10.00 a.m. to 5.00 p.m. the animal therefore received food on two separate occasions. The flow of digesta from cannula 1 under these conditions was always greater than that observed when the animal was fed once daily; thus in Fig. 4*B*, the flow was 885 ml./hr over the 7 hr of the experiment. The sheep ate 100 g oats and 700 g hay during this period.

Cannula 3 of sheep L 104 was placed about 10 cm caudal to the entrance into the duodenum of the common bile duct. Measurement of the flow of digesta from this cannula was therefore an index of the flow of digesta along the caudal part of the duodenum and differed from cannula 1 in that bile, pancreatic juice and duodenal secretion had been added to the digesta. In the experiment shown in Fig. 5*A*, done when the sheep was on once-daily feeding, the flow rate was 290 ml./hr over the experimental period of 7 hr, and in that shown in Fig. 5*B* 1026 ml./hr when the animal was fed three times daily.

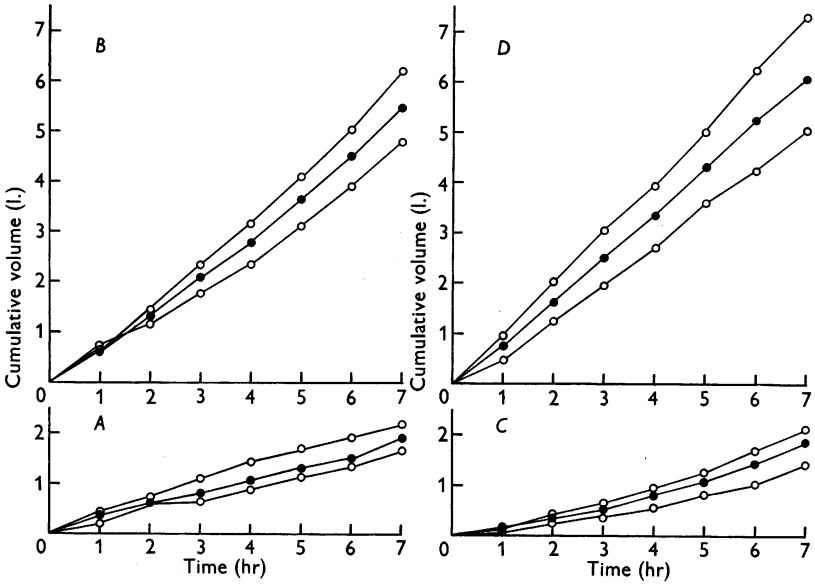

Fig. 6. Cumulative volumes of digesta obtained during 7 hr collection periods from: *A*, cannula 1 of sheep L 104 when fed once daily (3 expts.); *B*, cannula 1 of sheep L 104 when fed thrice daily (3 expts.); *C*, cannula 3 of sheep L 104 when fed once daily 3 expts.); *D*, cannula 3 of sheep L 104 when fed thrice daily (3 expts.). ●—● mean; ○—○ upper and lower limits.

The mean flows observed during three experiments on each parameter are shown in Fig. 6. The results represent mean values of 273 ml./hr and 785 ml./hr from cannula 1 when fed once and three times daily respectively and 263 and 864 ml./hr from cannula 3 when fed once and three times daily, respectively. The duration of all these experiments was 7 hr. In the experiments on the flow from cannula 1 the total volume of digesta collected over the whole period was measured and included in the calculations of mean flow rate. It was noticeable, however, that when the cap was first removed from cannula 1 there was frequently an immediate large

gush of digesta, which presumably represented drainage from the abomasum. Phillipson (1952) has also mentioned this occurrence and he omitted this material in his estimations of the mean hourly flow.

The reaction of the duodenal contents

During the experiments on the flow of digesta along the duodenum, the pH of each sample of digesta was determined. When fed once daily the pH of the digesta from cannula 1 was 2·70 ± 0·02 (S.E. of mean, 104 readings). In the experiments done when the sheep was used to thrice-daily feeding the pH was 2·70 ± 0·014 (265 readings). The corresponding figures for digesta from cannula 3 were: once-daily feeding, pH 4·10 ± 0·06 (102 readings); thrice-daily feeding, pH 3·70 ± 0·03 (256 readings) (Fig. 7).

It was possible, however, that the method of collection of digesta in the flow experiments might have given rise to erroneous pH values. Thus

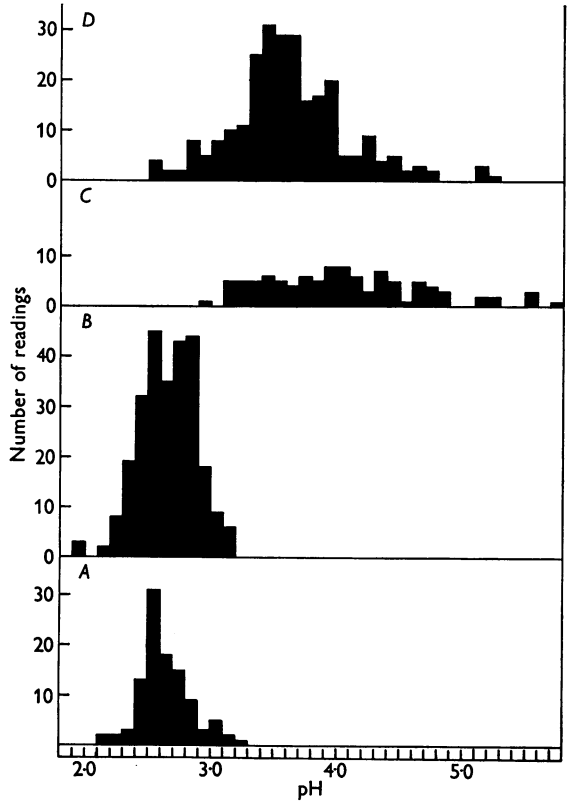

Fig. 7. The distribution of pH values of digesta obtained from the duodenal re-entrant fistulae. Sheep L 104. *A*, from cannula 1 when fed once daily; *B*, from cannula 1 when fed thrice daily; *C*, from cannula 3 when fed once daily; *D*, from cannula 3 when fed thrice daily.

thorough admixture of the bile and pancreatic juice with the digesta might have been prevented and the mixing effect of retrograde flow of digesta eliminated (Singleton, 1961). This point was investigated in one experiment by a less disruptive procedure in which the caps from cannula 1 and cannula 3 were removed momentarily and small quantities of digesta obtained for pH and acidity estimations. The pH values of the contents

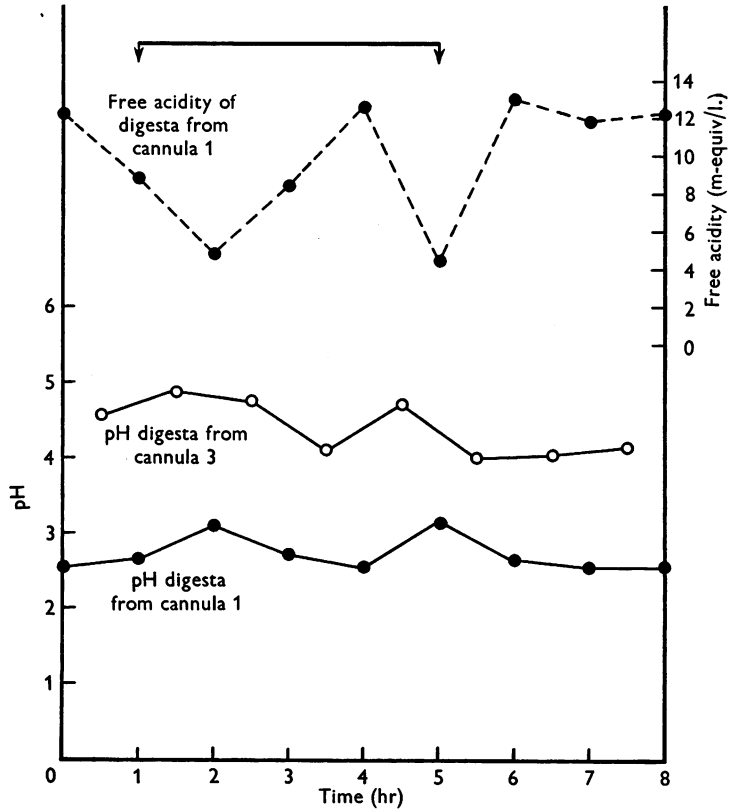

Fig. 8. The pH and free acidity of digesta obtained from cannulae 1 (●) and 3 (○) of the duodenal re-entrant fistulae by momentary interruption of flow of digesta. Sheep L 104. During period shown by top bar, 1000 g hay + 200 g oats fed.

from cannula 1 compared closely with those obtained in the flow studies but the pH of those from cannula 3, 4·40 ± 0·06 (8 obs.), was usually higher. The free acidity of the digesta from cannula 1 fluctuated after feeding and was accompanied by reciprocal changes in pH (Fig. 8).

Similar experiments were done on a sheep with cannulae placed in the duodenum about 30 cm from the pyloric sphincter and in the jejunum approximately 270 cm from the pyloric sphincter. The duodenal contents

were again of higher pH than those recorded in the flow studies and, as in the previous experiment, the increased secretion of gastric juice following a meal was reflected by a reduction in the pH of the contents obtained from both the duodenal and jejunal cannulae (Fig. 9). The increased acidity of the material leaving the abomasum after a single meal is evident from Fig. 10, where the pH and free acidity of the digesta obtained from a fundic abomasal fistula are represented alongside the secretory response observed from a gastric pouch prepared in the same animal.

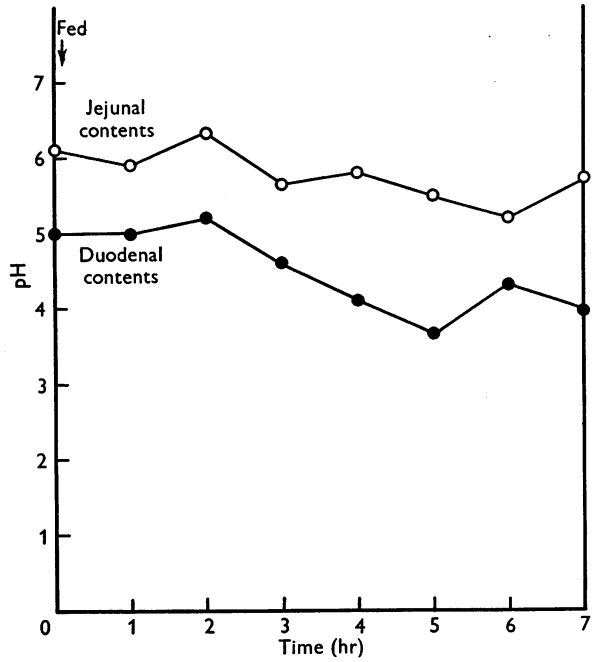

Fig. 9. The pH of duodenal (●) and jejunal (○) contents obtained by momentary interruption of flow of digesta before and after feeding: at arrow 700 g hay + 300 g oats fed.

Both these experiments were performed when the animals received one meal daily, conditions that are known to give rise to a typical gastric secretory response. Increasing the frequency of feeding so as to simulate natural feeding habits leads to more uniform secretory activity by the abomasum (Hill, 1960) and, from the results of determinations made on abomasal contents collected under these conditions, to a somewhat more uniform acidity of the digesta.

It is worth noting, however, that in some of the experiments done during three-times-daily feeding, in which small samples of digesta were removed from cannulae 1 and 3 without disrupting the flow, an occasional sample

contained a large amount of bile and the pH increased momentarily to between 5·0 and 6·0. The relatively greater flow of the mixed bile and pancreatic juice under these feeding conditions may have been responsible for this occurrence.

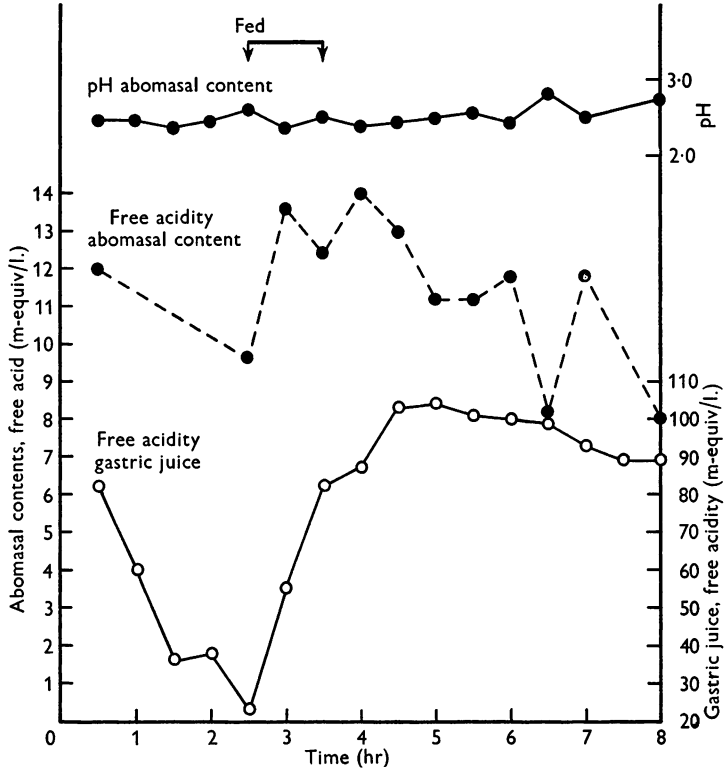

Fig. 10. The effect of a meal on free acid secretion by a gastric pouch and on the pH and free acidity of abomasal contents. During period shown by top bar, 325 g hay +200 g oats +425 g kale fed.

The digestive secretions and the pH of the duodenal digesta

The acid chyme which leaves the simple stomach is partially neutralized by the bile and pyloric, duodenal and pancreatic secretions. In contrast, the duodenal contents of the sheep are usually markedly acid and it would appear that the neutralizing capacities of the secretions are insufficient to deal with the relatively large volume of digesta which leaves the abomasum.

Experiments on a sheep with a gastric pouch prepared from the pyloric region of the abomasum showed that there was continuous secretion of a clear viscous juice which contained strands of visible mucus and clumps of desquamated epithelial cells. The juice was of pH 7·5–9·0 and all samples

exhibited proteolytic activity when assayed at pH 2·1, although this was much less than that of the juice obtained from fundic pouches. Hourly secretion varied from 5·0 to 12·0 ml. and there was an indication of slightly increased secretion after feeding. The average volume secreted during three experiments lasting altogether 18 hr was 8·0 ml./hr, and as the area occupied by the pouch was assessed to be about one third of the total pyloric region, the total daily volume of secretion was calculated to be about 500 ml. 42 ml. 0·1 N-HCl was required to reduce 100 ml. pyloric juice to pH 2·0–2·5; that is, approximately 200 ml. of 0·1 N-HCl was sufficient to bring the total daily output of pyloric juice to this level of acidity. As the estimated total daily output of acid gastric juice is 5·0 l. (Masson & Phillipson, 1952; Hill, 1960), the pyloric secretion appears to be of little significance in so far as neutralization of the abomasal content is concerned.

The difference in the pH of the digesta obtained from cannula 1 and cannula 3 of sheep L104 was attributed to the addition of duodenal secretion and the mixed bile and pancreatic juice. The respective roles of these secretions were demonstrated in one experiment in which samples of digesta were obtained from cannula 3 of sheep L104, by momentarily detaching the connexion with cannula 4. When bile and pancreatic juice were excluded from the digesta by catheterization, the mean pH over 14 hr was $2{\cdot}94 \pm 0{\cdot}09$ (14 obs.). When the catheter was removed and bile and pancreatic juice allowed to flow into the duodenum and mix with the digesta, the mean pH over 17 hr was $3{\cdot}28 \pm 0{\cdot}10$ (17 obs.).

DISCUSSION

The considerable development of the Brunner's glands and the observation that they extended along the duodenum as far as the entrance of the common bile duct confirmed the generally held view that these glands are more abundant in the herbivore than the carnivore (see Grossman, 1958). As the duodenal loop preparation included the whole of the Brunner's-gland area, the secretion collected from the isolated loop, when the common bile duct was catheterized, was a measure of the total Brunner's secretion plus that of the mucous epithelial cells lining the loop. There was no way of determining the volume of the latter but it was probably small compared with that of Brunner's secretion.

A more serious disadvantage inherent in the use of this preparation for studies on the secretion of Brunner's glands was the absence of the stimulatory effect of acid digesta in the loop during collection of the secretion. The average flow rate of 13·3 ml./hr was therefore a minimum one and may have merely represented spontaneous secretion; the output of Brunner's glands during the passage of digesta through the duodenum is

likely to be much greater. Florey & Harding (1934) obtained a comparable flow rate of 1–2 ml./hr from a 5 cm blind segment of goat duodenum when the animal fasted. In their preparation, however, acid digesta came into contact with some of the duodenal mucosa and the observed increase in secretion to 5·0 ml./hr after feeding was presumably caused by the liberation of secretin. The increased secretion obtained in the present experiments was unexpected since, despite the concomitant increase in the flow of digesta, these were not allowed to enter the duodenum during actual experiments. It is possible that increased movement of the tract may have stimulated secretion but the liberation of a secretogogue from some other region of the tract cannot be entirely ruled out.

Measurement of the total production of Brunner's secretion during the flow of digesta through the duodenum has still to be made, but from the relatively small change in acidity of the duodenal content when bile and pancreatic juice were excluded it is evident that, even if there is a fivefold increase in secretion from that observed here, the neutralizing role of Brunner's secretion is of little consequence. This deficiency in the buffering capacity of ruminant duodenal secretion has been noted previously by Havard (1934).

There is no information on the rate of cell replacement in the ruminant digestive tract, but it is well established that the epithelia of other animals undergo continuous renewal, particularly in the stomach and small intestine (Stevens Hooper, 1956). Bertalanffy (1960) has incriminated the acidity and proteolytic activity of the digesta in the desquamatory process and as there is no interdigestive resting phase in the ruminant a high rate of cell turnover is to be expected. The heavy deposit of cellular debris found in the duodenal secretion was in accord with this idea.

The validity of the results on the flow of the mixed bile and pancreatic juice depends on how little catheterization interferes with the normal secretory process. Insertion of a semi-rigid catheter through the sphincter of Oddi and into the common bile duct presumably eliminates sphincter action, and although there is no information on the activity of the sphincter in the sheep direct observation has shown that the bile and pancreatic juice escape into the duodenum in a series of spurts. This intermittent flow is converted to a continuous one by catheterization but as the gushes normally occur at frequent intervals it seems unlikely that the rate of secretion is greatly affected by elimination of sphincter activity. It is probable too, that catheterization results in drainage of the gall-bladder and elimination of gall-bladder function. The absorptive capacity of the ruminant gall-bladder is, however, generally assumed to be small.

Non-return of pancreatic juice to the duodenum does not affect the rate of secretion of the juice in the sheep (Taylor, 1958) but, as in other species,

it is essential if bile secretion is to be maintained at its normal level that there should be no loss of bile from the animal. The results of experiments in which the mixed secretion was collected and not returned to the duodenum are therefore of little value as an indication of the normal rate of secretion. However, they indicate the minimum output of mixed secretion and when this is compared with that of pure bile obtained under similar conditions it appears that approximately equal proportions of bile and pancreatic juice are present in the mixed secretion. Under natural conditions of uninterrupted re-circulation of bile salts it is likely that there will be a greater proportion of bile in the mixed secretion.

The close relationship between the flow of digesta and the secretion of bile and pancreatic juice was demonstrated by the increase in secretion which occurred when the ration was given on three separate occasions during the day. This procedure caused an increased flow of digesta and, although the total daily secretion of mixed bile and pancreatic juice was not determined under these conditions, it was evident that within the 12 hr experimental period the augmented flow of digesta was associated with an increased secretion of mixed bile and pancreatic juice. Whether there was an increased secretion of one or both components was not determined.

The factors which regulate the secretion of gastric juice by the abomasum and the acidity of the abomasal content have been discussed recently by Hill (1960) and Ash (1961*a*, *b*), and it is evident that the primary stimulus to gastric secretion is the flow of digesta from the forestomachs to the abomasum. Regulatory mechanisms appear to co-ordinate the two processes so that the digesta which leave the abomasum have a comparatively uniform reaction. Nevertheless, the secretory response to a single meal, after a fast, observed as a marked increase in acid secretion from an isolated gastric pouch, was also evidenced by increased acidity of the digesta as they passed down the intestine. Increasing the frequency of feeding, although not the total amount of food consumed, smoothed out, to some extent, the fluctuations in acidity. During natural grazing, where feeding is a more frequent process, there may be an even more uniform flow of digesta of constant acidity. This does not preclude rapid changes in abomasal secretory activity and in the reaction of the duodenal contents which may occur with a sudden change of diet.

The low pH of the duodenal digesta, even when collected caudal to the entrance of the common bile duct, showed that the bile and pyloric, duodenal and pancreatic secretions had little neutralizing effect on the acid abomasal digesta. Magee (1961) has also commented on the high acidity of the duodenal contents of the sheep and on the problem of how the pancreatic enzymes function in this environment. In this respect it would seem reasonable to consider the duodenum as a continuation of the abo-

masum where peptic activity can continue and to expect that pancreatic digestion may occur further along the intestine.

The essential continuity of the flow of digesta in the ruminant duodenum has been emphasized by Hogan & Phillipson (1960) and Singleton (1961), who have measured the flow in this region though they made no attempt to relate this to the frequency of feeding. The purpose of the present experiments was to study the relationships between flow of digesta and the secretory activity of the digestive glands over short periods, and hence no information was obtained on the total daily flow. Nevertheless, on an hourly basis, the flows of 360 ml. (Hogan & Phillipson, 1960) and 458 ml. (Singleton, 1961) obtained from sheep fed twice daily are intermediate between the figures reported here for once- and thrice-daily feeding. It would appear therefore that frequency of feeding and rate of flow of digesta are related and that the precise feeding and watering routine should be indicated in any studies on flow of digesta. In this respect it may also be important to relate the actual time spent in eating and ruminating to flow measurements, since there was an indication in some of our experiments that these activities were accompanied by an increased flow.

It was originally expected that the addition of the digestive secretions to the duodenal contents would result in a greater flow of digesta from cannula 3 of the duodenal re-entrant fistula compared with that from cannula 1, and therefore provide an indication of the volume of secretions added during the passage of digesta through the duodenum. A comparison of the mean flow obtained from cannula 1 (785 ml./hr; 3 experiments) with that obtained from cannula 3 in separate experiments (864 ml./hr; 3 experiments) when the animal was fed three times daily indicated that the total volume of secretion added was 79 ml./hr. However, there was no significant difference between the flow of digesta from cannula 1 (273 ml./hr) and cannula 3 (263 ml./hr) in separate experiments when the animal was fed once daily. The reason for this discrepancy is not clear and this point is being investigated by simultaneous measurement of the flows from cannula 1 and cannula 3. In the one experiment so far completed with the sheep on once-daily feeding, the mean flow of digesta from cannula 1 over the 7 hr period was 279 ml./hr and from cannula 3, 323 ml./hr. A mean volume of 44 ml./hr of bile, pancreatic juice and duodenal secretion was therefore added to the digesta during their passage through the duodenum.

Although more information is required on both the flow of digesta and the behaviour of the secretory glands under different feeding conditions, the results presented here indicate that the two are closely related. In this respect the composition and physical nature of the diet may also be important.

SUMMARY

1. Measurements have been made of the flow of digesta and digestive secretions in the duodenum of the conscious sheep.

2. Brunner's glands were found to extend from the pylorus to the sphincter of Oddi and were included in a duodenal loop preparation which could be isolated for the collection of pure secretion. In the absence from the loop of digesta, the secretory rate varied from 13·3 to 26·0 ml./hr.

3. Mixed bile and pancreatic juice were collected by catheterization of the common bile duct. There was a large initial volume, attributable to drainage of the gall-bladder, followed by a decline in secretory rate associated with the interruption of the entero-hepatic circulation of bile salts. The average secretory rate during the 4–24 hr period of collection was 38·8 ml./hr or 0·74 ml./kg/hr.

4. Comparable experiments on other sheep showed that the average secretion of bile only during the 4–24 hr period was relatively constant at 16·8 ml./hr or 0·41 ml./kg/hr.

5. A change in the frequency of feeding from once to three times daily and the return of secretion to the duodenum both caused an increase in secretion of the mixed bile and pancreatic juice.

6. Pyloric secretion collected from an isolated pouch had relatively poor neutralizing properties and the total daily secretion was estimated to be about 500 ml.

7. The flow of digesta from the abomasum was increased from 273 ml./hr when the animal was fed once daily to 785 ml./hr when the same ration was given in three feeds. Likewise the flow along the duodenum was increased from 263 to 864 ml./hr.

8. The duodenal digesta were markedly acid even after the addition of duodenal secretion, pancreatic juice and bile.

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