J. Physiol. (1982), **326**, *pp.* 453–459
With 5 text-figures
Printed in Great Britain

THE ROLE OF GASTRIC SECRETION IN POST-DIVERTED PANCREATIC HYPERSECRETION IN CONSCIOUS RATS

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(*Received 11 July 1979*)

SUMMARY

1. In rats prepared with chronic external pancreatic fistulae, gastric fistulae and choledocho-enterostomy, the volume and protein secretion of pancreatic juice was measured.

2. The pancreatic secretion of water and protein after juice diversion was significantly lower when the gastric fistula was open than when closed. Secretion was abolished after atropine, with the fistula open and in animals with ligated pylori.

3. Intraduodenal HCl significantly raised juice volume and protein in animals with diverted juice when the gastric fistula was open.

4. Soybean trypsin inhibitor produced a significantly smaller elevation of juice volume and protein when the gastric fistula was open.

5. It is concluded that the initial hypersecretion of pancreatic juice and protein, following its diversion from the duodenum, is a consequence of the entry of gastric juice into the intestine.

INTRODUCTION

Diversion of pancreatic juice from the duodenum in rats results in a rapid augmentation of pancreatic protein and water secretion (Green & Lyman, 1972; Green, Olds, Matthews & Lyman, 1973; Petersen & Grossman, 1977). It has been suggested that the presence of trypsin in the duodenum depresses the responsiveness of mucosa to stimulants capable of releasing secretin and/or cholecystokinin-pancreozymin (CCK) (Green & Lyman, 1972; Green *et al.* 1973; Schneeman & Lyman, 1975). This view is supported by the finding that fresh pancreatic juice placed in the duodenum will return the pancreatic hypersecretion of juice diversion to normal in rats (Green & Lyman, 1972; Green *et al.* 1973; Petersen & Grossman, 1977) and depress normal secretion in pigs (Corring, 1974). This response is not seen in juice which has been boiled (Hong, Nakamura & Magee, 1967). In addition, soybean

Owing to an error in the Press office, for which we apologize, we regret that the appearance of this paper has been delayed.

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trypsin inhibitor placed in the duodenum of normal animals produces a rapid augmentation of juice volume and its contained protein as does juice diversion to the exterior (Green & Lyman, 1972; Green *et al.* 1973; Schneeman & Lyman, 1975). The purpose of the present study was to assess the role, if any, of endogenous gastric acid in mediating this response.

METHODS

Surgical procedure. Male Sprague Dawley rats weighing 355–485 g were operated upon, after a 12 hr fast, under anaesthesia in order to provide pancreatic and duodenal fistulae and internal choledochoduodenal fistulae (Demol & Sarles, 1978). Each animal was provided with a large stoppered stainless steel gastric fistula (i.d. 4 mm) which connected the glandular stomach to the exterior through the left anterior abdominal wall. In five animals the pylorus was ligated. After operation, all animals were placed in Bollman restraining cages, where they remained until the experiments were complete. Convalescence lasted 3 days during which time the pancreatic and duodenal cannulae were connected, the gastric cannula closed, and the animals allowed food and water *ad libitum*.

Experimental protocol. Rats having a ligated pylorus and open gastric fistula received no food and water by month. They were given glucose Ringer by vein continuously (1 ml./hr) and used for experiment after 36 hr. All of these showed an increase in pancreatic juice volume and protein output in response to intraduodenal HCl.

In all the other animals, experiments were carried out on the fourth to seventh day after surgery. These animals were deprived of food but not water for 12 hr before experiment. Water was removed on the day of experiment and an intravenous saline infusion started (1 ml./hr). The pancreatic cannula was disconnected from the duodenal cannula. Previously collected rat pancreatic juice was then infused into the duodenum (0·6 ml./hr) and the basal secretion collected from the pancreatic cannula for three successive 30 min collection periods. The duodenal infusion of pancreatic juice was then replaced by saline or 0·05 N-HCl to study juice diversion secretion. The effects of atropine injection (I.V. 300 μg/kg . hr), soybean trypsin inhibitor (20 mg; Merck, art. 24020) and pancreatic juice (1·2 ml.) on diversion secretion were also measured.

The above procedures were always conducted with the gastric fistula either open or closed so that comparisons could be made on the same animal. The only exception to this was in the case of the pylorus ligated animals in which the effects of juice diversion and HCl were studied. Experiments carried out with gastric fistula open or closed took place on separate days. The sequence for carrying out these procedures was random.

Chemical determinations. Protein concentrations were determined by absorption at 280 nm ($E^{1\%}_{1cm} = 20$) and the acid concentration of collected gastric juice was estimated by titration against 0·02 N-NaOH.

Statistical analysis. Statistical significance was evaluated by application of the paired t test to the differences seen between increases with the fistula open and closed, with and without atropine or HCl present ($P < 0·05$ was taken as level of significance).

RESULTS

Diversion of pancreatic juice

Diversion of juice from the duodenum with the gastric fistula closed resulted in the often described pattern of augmented water and protein secretion (Fig. 1). When the gastric fistula was open, however, the rapid augmentation to the first peak was not seen. This was especially evident in the case of the protein output. Throughout the 4 hr following juice diversion there was a slow increase in juice volume and protein which, however, did not reach the levels seen when the gastric fistula was shut.

The gastric acid collected from the fasted rat (initially 100–130 μequiv/30 min) fell

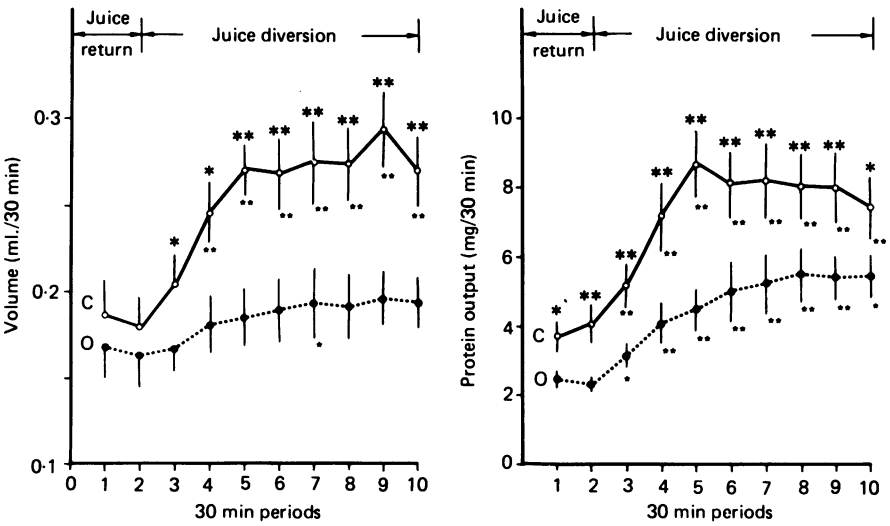

Fig. 1. Mean effect ± S.E. of the mean of diverting pancreatic juice from the intestine on pancreatic water (left) and protein secretion (right) with the gastric fistula closed (C) and open (O). *, ** Difference between C and O significant at 95 and 99% levels, respectively. *, ** Difference from pre-diversion control significant at 95 and 99% levels, respectively. $n = 11$ rats.

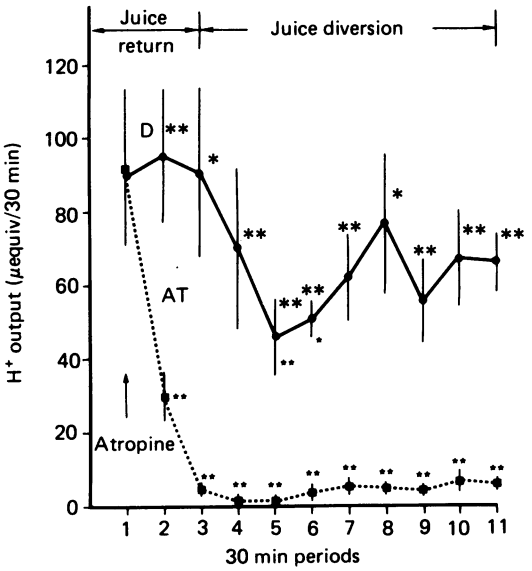

Fig. 2. Mean effect ± S.E. of the mean of atropine on gastric acid secretion (AT) after diversion of pancreatic juice. D: gastric fistula open without atropine as control. *, ** Difference between AT and D significant at 98 and 99% levels, respectively. *, ** Difference from pre-atropine or pre-diversion control significant at 98 and 99% levels, respectively. $n = 7$ rats.

to a lower plateau (about 70 μequiv/30 min after 2 hr) whether pancreatic juice was diverted or not. Basal secretion of pancreatic water and protein was low ($0{\cdot}07 \pm 0{\cdot}02$ (S.E. of the mean) ml./30 min; $0{\cdot}06 \pm 0{\cdot}03$ mg/30 min, respectively). Diversion of pancreatic juice from the duodenum produced no significant increase in either of these rates of secretion.

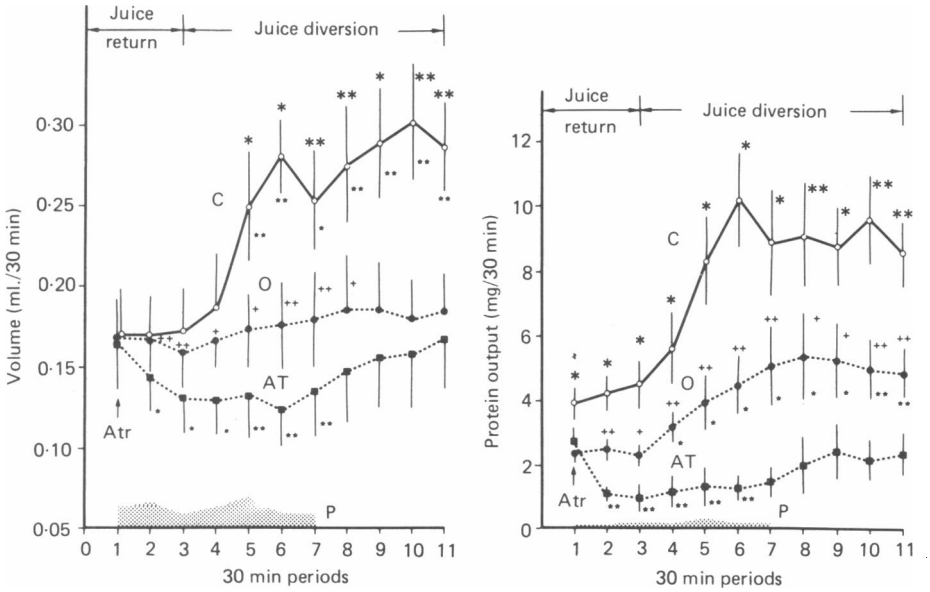

Fig. 3. Mean effect $\pm$ S.E. of the mean of atropine (Atr) in animals with open gastric fistulae (AT) and of diverting pancreatic juice in animals with closed gastric fistulae (C), open gastric fistulae (O) and ligated pylori (P) on pancreatic water (left) and protein secretion (right). *, ** Difference between C and O significant at 95 and 99% levels, respectively. +, ++ Difference between O and AT significant at 95 and 99% levels, respectively. *, ** Difference from pre-atropine or pre-diversion control significant at 95 and 99% levels, respectively. $n = 7$ rats.

Atropine

Atropine reduced gastric acid secretion almost to zero (Fig. 2). It also caused a pronounced diminution of post-diversion pancreatic secretion of protein and water (Fig. 3).

Intraduodenal HCl

In animals with open gastric fistulae and pancreatic juice diversion 0·05 N-HCl introduced into the duodenum at a rate comparable to that of the gastric acid secretion (1·2 ml./hr, 60 μequiv/hr) caused a significant increase in both juice volume and protein secretion (Fig. 4). In the pylorus ligated animals intraduodenal HCl produced a small but significant increase in protein secretion ($0{\cdot}43 \pm 0{\cdot}12$ mg/30 min, $P < 0{\cdot}05$).

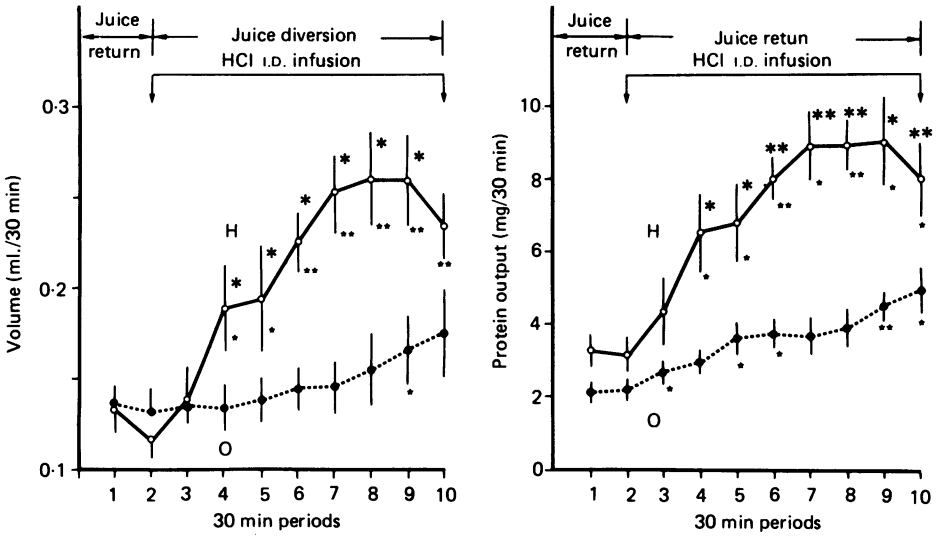

Fig. 4. Mean effect ± S.E. of the mean of intraduodenal (I.D.) infusion of HCl (60 μequiv/hr) on pancreatic water (left) and protein secretion (right) in animals with open gastric fistulae and diverted pancreatic juice (H). O: gastric fistula open without HCl infusion as control. *, ** Difference between H and O significant at 95 and 99% levels, respectively. *, ** Difference from pre-diversion control significant at 95 and 99% levels, respectively. $n = 5$ rats.

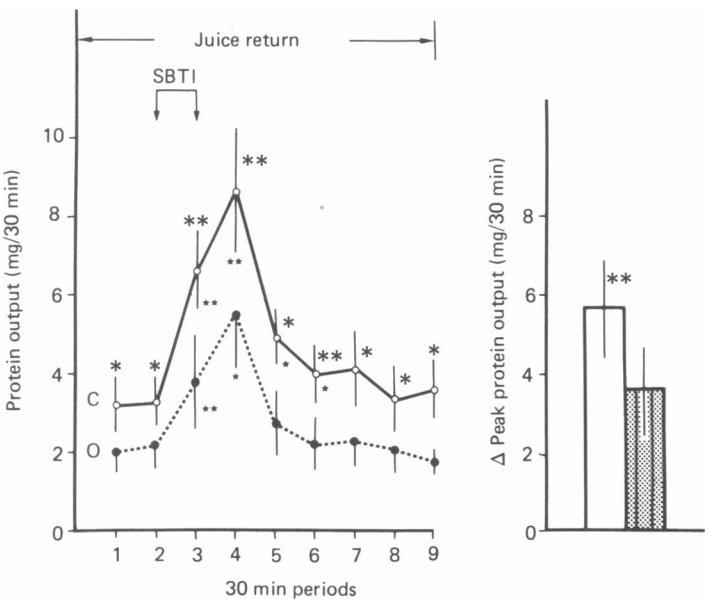

Fig. 5. Mean effect ± S.E. of the mean of intraduodenal infusion of soybean trypsin inhibitor (SBTI) (10 mg/30 min per rat) in animals with closed (C) and open (O) gastric fistulae on pancreatic protein secretion (left) and a comparison of the peak protein increase (right). *, ** Difference between C and O significant at 95 and 99% levels, respectively. *, ** Difference from pre-SBTI control significant at 95 and 99% levels, respectively. $n = 8$ rats.

Trypsin inhibitor

Intraduodenal soybean trypsin inhibitor caused a much smaller augmentation of juice protein when the gastric fistula was open and gastric acid diverted to the exterior (Fig. 5). The augmentation in volume in two experiments was significantly lower when the gastric fistula was open than when closed in the second sample after infusion of the inhibitor. Trypsin inhibitor did not significantly alter the recovery of gastric acid.

DISCUSSION

The resting gastric secretion in the rat was substantial. The rapid fall on opening the gastric fistula, which was seen whether or not the pancreatic juice was diverted, suggests that secretion was much higher with the fistula closed than the plateau attained with the fistula open. Our results indicate that this gastric juice contributed substantially to the hypersecretion seen following diversion of pancreatic juice from the duodenum. This hypersecretion might result from increased acidity in the duodenum, due to the absence of pancreatic bicarbonate, or from an increased sensitivity of receptors resulting from the absence of trypsin or from a mixture of both of these effects. In either case diversion of gastric contents to the exterior should reduce post-diversion hypersecretion. Present results show that such a reduction does indeed take place. Earlier workers have failed to increase the post-diversion hypersecretion further by intraduodenal infusion of HCl (Grossman, 1958; De Smul, De Waele, Wissocq & Kiekens, 1974). Duodenal receptors in those animals could have been maximally stimulated by gastric contents, especially acid. We have found that the lower rates of secretion found in animals with open gastric fistulae (protein and water) are easily increased by intraduodenal HCl.

The reason secretion took place at all when the gastric fistula was open is probably because gastric contents still continued to enter the duodenum. This view is supported by experiments in the animals with ligated pylori, where gastric juice was excluded from the duodenum. In this case basal pancreatic secretion was small and not increased by juice diversion. The pancreata of these animals were, however, responsive given adequate stimulation by HCl.

Experiments with atropine, in which acid secretion was almost abolished and pancreatic secretion was low, also support this hypothesis. Using the open gastric fistula and the atropine experiments and plotting acid recovered against pancreatic protein at the time of its maximum response (Figs. 2 and 3, sample 6), a highly significant correlation was obtained ($r = 0{\cdot}744$, $P < 0{\cdot}01$, $n = 14$). It is reasonable to assume that acid entry into the duodenum is directly related to the level of acid secreted. These results do not agree with those of De Waele, De Smul, Wissocq & Keikens (1974). The reason for this remains unknown.

If trypsin inhibitor itself stimulates CCK receptors in the duodenum, as suggested by Green *et al.* (1973), its effect should be uninfluenced by opening or closing the gastric fistula. Its effect was, however, much smaller on protein and volume secretion when gastric juice was diverted. The likely explanation for this is that trypsin depresses the sensitivity of duodenal receptors which respond to gastric contents. These receptors are unblocked in the trypsin inhibitor exeriments with the gastric

fistula open, but there is less stimulus entering from the stomach under these conditions. If this view is correct, one would expect no trypsin inhibitor-induced augmentation of pancreatic secretion if gastric contents were completely diverted from the duodenum. This experiment has yet to be carried out.

It is concluded that the early phase of pancreatic hypersecretion following diversion of juice from the duodenum in the rat is due to maximal or near maximal stimulation of duodenal receptors by basal gastric acid secretion which in this animal is very high. The pig, also, has a very high basal gastric acid secretion which may explain why its responses to trypsin inhibitor and intraduodenal pancreatic juice resemble those in the rat and not those of dogs, in which basal gastric acid secretion is low or absent.

Our results with trypsin inhibitor favour the view that pancreatic trypsin depresses duodenal secretin and/or CCK-PZ receptors, but the present experiments do not clarify the role, if any, of bicarbonate.

We wish to thank Georges Michel and Marie-Alix Devaux for their help of the experiments.

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