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EFFECT OF PANCREATIC JUICE ON BASAL PANCREATIC AND GASTRIC SECRETION IN DOGS

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SUMMARY

1. The effect of duodenal infusion of pancreatic juice on basal pancreatic and gastric secretion was studied in five conscious dogs provided with pancreatic fistulae, gastric fistulae and Heidenhain fundic pouches.

2. Pancreatic juice and trypsin stimulated a pancreatic secretion rich in protein.

3. Autodigested juice without proteolytic activities also stimulated the secretion. Boiling the juice or addition of trypsin inhibitor to the juice diminished the augmented secretion.

4. It seems, therefore, that trypsin is necessary even in proteolytically inactive autodigested juice for pancreatic stimulation.

5. In dogs, unlike rats and pigs, basal pancreatic secretion is not under negative feed-back control by duodenal tryptic activity.

6. Basal gastric secretion was not significantly changed by duodenal infusion of pancreatic juice.

INTRODUCTION

Diversion of pancreatic juice from the duodenum in the rat results in a rapid augmentation of pancreatic secretion (Green & Lyman, 1972; Green, Olds, Matthews & Lyman, 1973; Schneeman & Lyman, 1975; Peterson & Grossman, 1977). Return of the juice into the duodenum reduces the secretion to the original level. This phenomena has been observed in pigs (Magee & Hong, 1965; Hong, Nakamura & Magee, 1967; Corring, 1973; Ihse & Lilja, 1979). Trypsin has the same effect as pancreatic juice and its inhibitor augments the pancreatic secretion when the juice is returned into the duodenum (Green & Lyman, 1972; Green *et al.* 1973; Schneeman & Lyman, 1975; Ihse & Lilja, 1979). From these observations it has been proposed that in these species pancreatic secretion receives a negative feed-back inhibition activated by duodenal trypsin (Green & Lyman, 1972; Green *et al.* 1973; Schneeman & Lyman, 1975; Corring, 1973; Ihse & Lilja, 1979).

Recently Noda, Magee & Sarles (1982) observed that this hypersecretion caused by juice diversion in rats is greatly diminished by diverting the gastric juice. In the present study the role of the duodenal pancreatic juice on basal pancreatic and

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gastric secretions was studied in dogs, in which, unlike rats, basal gastric secretion is very low.

METHODS

Five mongrel dogs weighing 18–27 kg were provided with Thomas duodenal fistulae, gastric fistulae and Heidenhain fundic pouches. The experiments were started one month after the operation with a week interval between tests.

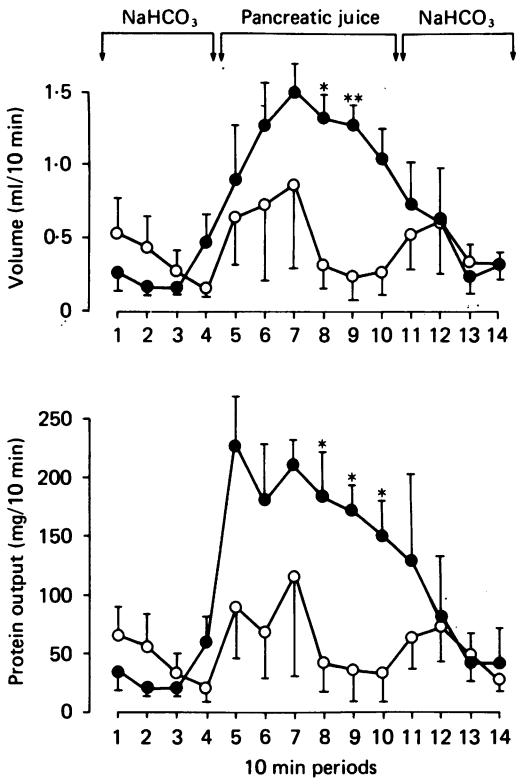

Fig. 1. The effect of duodenal infusion of pancreatic juice on basal pancreatic secretion. Filled and open circles show the secretion (mean ± S.E. of mean) in response to pancreatic juice and control ($NaHCO_3$), respectively. Asterisks indicate significant differences (*$P < 0.05$, **$P < 0.01$) from control. $n = 5$.

The animals were held in Pavlov stands after an 18 h fast. Collections of secretion were made at 10 min intervals. Pancreatic secretion was collected by cannulation of the duct, gastric fistula secretion by drainage, and pouch secretion by the saline washout method (Magee & Nakajima, 1968).

Duodenal infusion was made at 5 ml/10 min through a balloon catheter (Fr. 24) via the duodenal fistulae. Sodium bicarbonate solution (100 mM-$NaHCO_3$, 50 mM-NaCl) was used as a control and as the solvent for substances tested. After a 40 min control infusion the infusate was changed for the test solution for 60 min, followed by another 40 min control infusion.

The solutions tested were prepared as follows.

Pancreatic juice. Pancreatic juice collected by direct cannulation from five dogs was pooled (about

1·8 l) and mixed well at 4 °C. It was then divided for each test (about 40 ml) and was kept at −20 °C before use. Bicarbonate and protein concentrations of the thawed juice were 110 m-equiv/l and 15 mg/ml, respectively.

Autodigested pancreatic juice. The above juice was incubated with enterokinase (Nutritional Biochemicals Company) at 40 °C for 5 d and then heated at 70 °C for 20 min. No proteolytic activities were found.

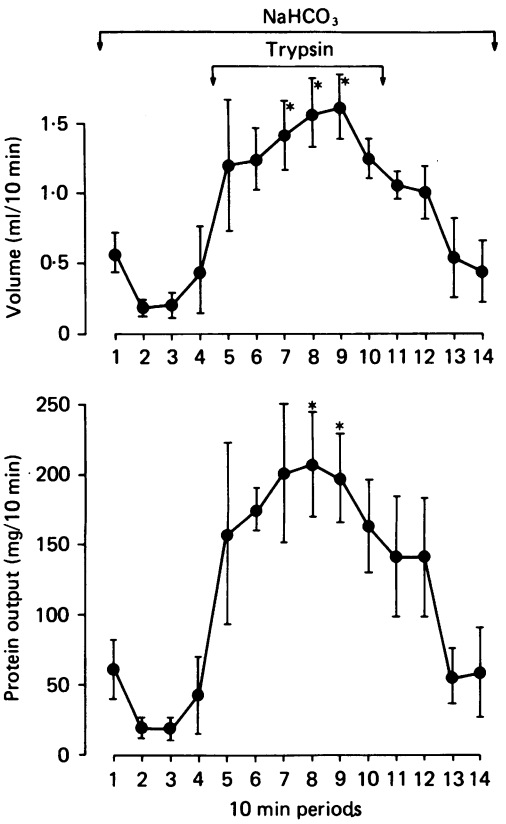

Fig. 2. Pancreatic secretion (mean ± S.E. of mean) in response to the duodenal infusion of trypsin (100 mg/10 min). Asterisks indicate significant differences ($P < 0·05$) from control. $n = 5$.

Boiled pancreatic juice. Frozen pancreatic juice above was thawed and then heated at 90 °C for 4 min in a waterbath before use.

Synthetic trypsin inhibitor. [*N*,*N*-dimethylcarbanoylmethyl 4-(4-guaninobenzoyloxy)-phenyl acetate] methansulphonate (FOY-305®) (kindly donated by Ono Pharmaceutical Company, Osaka, Japan) was dissolved in the control infusate (10^{-3} M). Proteolytic activities of 1 mg/ml crystalline trypsin (Boehringer Mannheim, West Germany) were completely inhibited by this inhibitor at 10^{-6} M, but those of 1 mg/ml crystalline chymotrypsin (Sigma type II) were not inhibited by up to 10^{-2} M inhibitor (Tamura, Hirado, Okumura, Minato & Fujii, 1977).

Pancreatic juice with synthetic trypsin inhibitor. No proteolytic activities were found in pancreatic juice with 10^{-3} M inhibitor.

Trypsin. Porcine trypsin 1–300 (United States Biochemical Company) was used (20 mg/ml).

Additional experiments were made with pancreatic juice activated by enterokinase. Total proteolytic activity of the juice was not changed with the addition of the inhibitor (10^{-3} M).

Acid was measured by titration with 50 mN-NaOH (Radiometer Titrator II). Bicarbonate concentration was estimated by titrating the acid which remained after heating the mixture of 0·5 ml of the sample and 1 ml of 0·1 N-HCl. Protein concentration was measured by spectrophotometry (Hitachi-Perkin Elmer) at 280 nm with bovine serum albumin (Sigma) as standard. Peptic and total proteolytic activities were estimated by the method of Anson (1938).

The mean values of both 10 min periods and 60 min periods (the sum of six 10 min period values) were compared with paired t test (two tailed). $P < 0.05$ was considered to be significantly different.

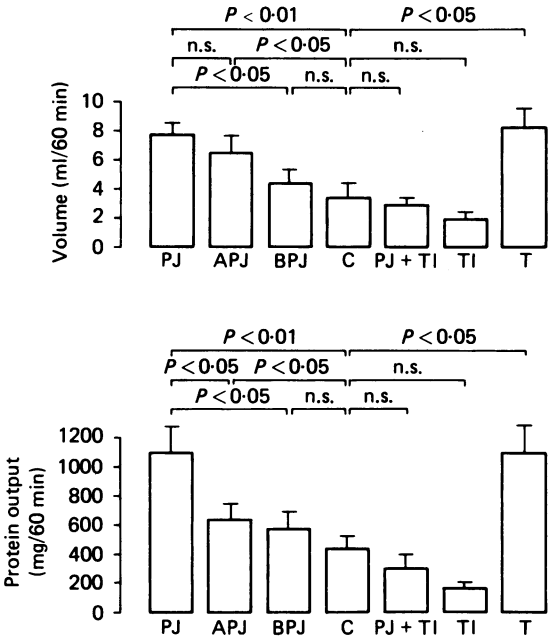

Fig. 3. The effect of duodenal additions on basal pancreatic secretion. Bars indicate the sum in 60 min (mean ± S.E. of mean). $n = 5$. PJ: pancreatic juice; APJ: autodigested pancreatic juice; BPJ: boiled pancreatic juice; C: control; TI: trypsin inhibitor; PJ + TI: pancreatic juice with trypsin inhibitor; T: trypsin; n.s.: not significant.

RESULTS

Duodenal infusion of pancreatic juice stimulates pancreatic water and protein secretion (Fig. 1). The effect appeared quickly with the appearance of dark coloured bile (in two of five dogs at period 5, in three at period 6, in four at period 7 and in all at period 8) from the duodenal fistula. Trypsin had the same effect as pancreatic juice (Fig. 2). Basal pancreatic secretion showed the periodic fluctuation, which resulted in wider variations in some control samples (Fig. 1).

The effects of various duodenal additions on 60 min cumulative secretion (the sum from periods 5 to 10) are shown in Fig. 3. The stimulatory action of pancreatic juice was diminished by boiling the juice or by the addition of trypsin inhibitor to it. Autodigested pancreatic juice without proteolytic activities stimulated the secretion, though less effectively than the native juice.

Trypsin inhibitor suppressed the augmented secretion caused by the activated

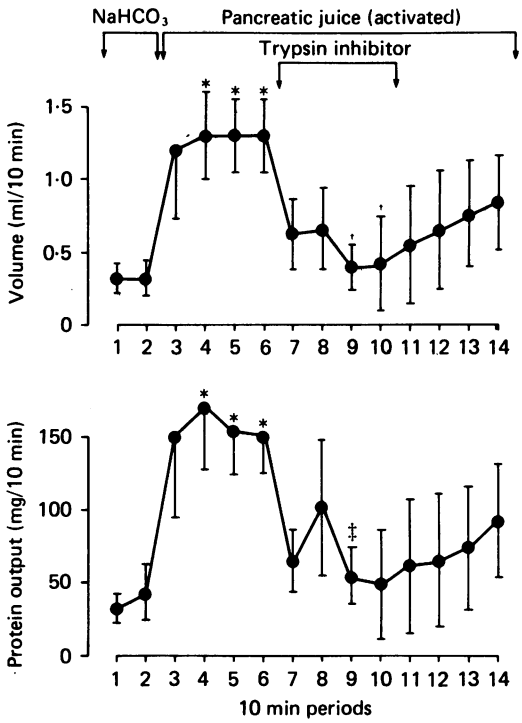

Fig. 4. The effect of trypsin inhibitor on pancreatic secretion stimulated by the activated pancreatic juice. Asterisks indicate the significant differences ($P < 0.05$) from control (the mean of sample 1 and 2). Daggers indicate the significant differences ($\dagger P < 0.05$, $\ddagger P < 0.01$) from the plateau value (the mean of sample 5 and 6). $n = 5$.

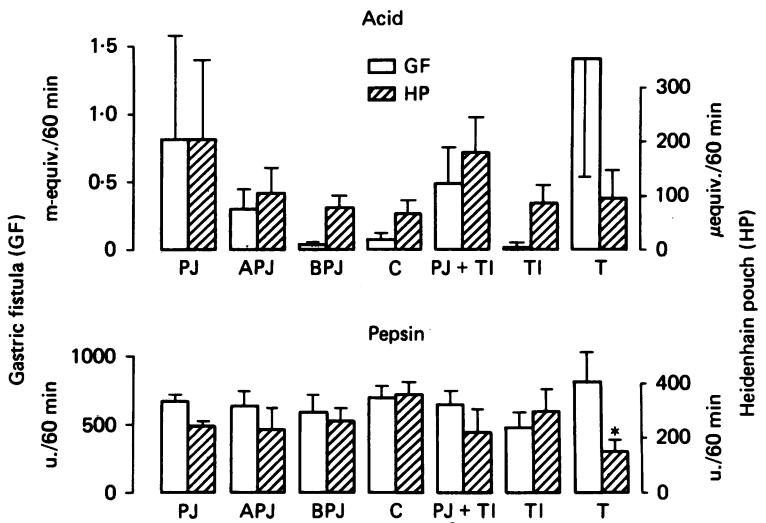

Fig. 5. The effect of duodenal additions on gastric secretion. Bars indicate the sum in 60 min (mean ± S.E. of mean). $n = 5$. See Fig. 3 for abbreviations.

pancreatic juice (Fig. 4). It also depressed the unstimulated secretion, but the difference from control was not significant (Fig. 3).

Fig. 5 shows the effects of duodenal additions on basal gastric secretion. Gastric secretion was not changed significantly by duodenal additions, except for the pouch pepsin secretion during trypsin infusion.

DISCUSSION

In dogs basal pancreatic secretion is not the hypersecretion resultant from the diversion of the juice, since infusion of the juice or continuous return of the juice into the duodenum fails to suppress it (Zucker, Newburger & Berg, 1932). Food-stimulated water and bicarbonate secretion, but not protein secretion, can be greatly diminished by returning the juice or bicarbonate solution (Annis & Hallenbeck, 1951; Cooke, Nahrwold & Grossman, 1967; Sale, Goldberg, Fawcett & Wormsley, 1977). This could be explained by the absence of the neutralizing effect of the juice after diversion, resulting in an enhanced activation of the secretion mechanism (Annis & Hallenbeck, 1951; Cooke *et al.* 1967).

Boldyreff (1911) first observed the periodical change of basal pancreatic secretion in the fasting dog. Fluctuation of basal secretion has been observed by others (Babkin & Ishikawa, 1912; Zucker *et al.* 1932; Scott, Graham & McCartney, 1940). In this study statistical analysis was made on the summed effects, which minimize the influence of the spontaneous changes in secretion (Elashoff, 1981).

A large number of substances given intraduodenally have been tested for their effects on pancreatic secretion in dogs (Thomas, 1950; Harper, 1967), but it seems few have studied the effect of the juice itself. Zucker *et al.* (1932) observed no inhibition of the secretion by the juice, while Hong *et al.* (1967) noted a depression. In the present study, pancreatic juice and trypsin stimulated a secretion rich in protein.

Tryptic activities seem to be important for this, since boiling the juice or addition of trypsin inhibitor to the juice diminished the effect. Furthermore, the augmented secretion caused by the activated juice was also suppressed by trypsin inhibitor even though its total proteolytic activity remained unreduced. However, autodigested juice without proteolytic activities stimulated the secretion. These suggest that stimulatory action of the juice is exerted by some part of the trypsin molecule regardless of its proteolytic activity, perhaps via activation of the cholecystokinin mechanism.

The action of pancreatic juice, trypsin and its inhibitor on basal pancreatic secretion in the dog seem to be the opposite of these in the rat and the pig (Green & Lyman, 1972; Green *et al.* 1973; Schneeman & Lyman, 1975; Magee & Hong, 1965; Hong *et al.* 1967; Corring, 1973; Ihse & Lilja, 1979). So far no conclusive results have been obtained in man (Ihse, Lilja & Lundquist, 1977; Kawisz, Miller, DiMagno & Co, 1980). The work of Noda *et al.* (1982) suggests that trypsin may not be the sole factor in duodenal feed-back mechanism even in species where such a mechanism is evident.

It has been known in dogs that the exclusion of pancreatic juice from the intestine either by pancreatotomy or ligation of pancreatic ducts leads to the gastric

hypersecretion (Steinberg, 1921; Fauley & Ivy, 1929). In the present study only pouch pepsin secretion was suppressed by duodenal infusion of trypsin. It seems that temporary diversion or infusion of the juice does not influence the gastric secretion (Elliott, Taft, Passaro & Zollinger, 1961; Greenlee, Nelsen & Dragstedt, 1961; Hein, Silen & Harper, 1963).

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