Vasoactive Intestinal Peptide: Secretin-Like Action on the Avian Pancreas

Recently, a vasoactive intestinal peptide (VIP) has been isolated from hog duodenum and shown to be related in structure to the hormones secretin and glucagon[1-3]. The physiological role played by VIP is uncertain, but this peptide is known to have vasodilator and hypotensive actions in the dog[1] and to be 5–10% as potent as porcine secretin in stimulating the flow of juice from the pancreas in the cat[2]. In the present study further information on the significance of the secretin-like actions of VIP was sought by investigating its effect on the pancreas in turkeys and rats.

Materials and methods. Young turkeys (0.5–4.0 kg) were anaesthetized with urethane (1.5–1.75 g/kg, i.p.). A wing vein was cannulated for injections, and a polythene catheter was inserted into the gizzard to drain the acid secretions. The dorsal pancreatic duct was ligatured and the ventral duct was cannulated with polythene tubing (Portex pp 10). Pancreatic juice was collected in calibrated capillary tubes and the rate of flow noted at 10 min intervals. Protein concentration in the juice was estimated from the optical density at 280 nm. For the studies on pancreatic secretion in rats, animals were anaesthetized with urethane and pancreatic juice was collected by the method described by DOCKRAY[4].

Results and discussion. In a previous study it was found that pure natural porcine secretin stimulated pancreatic secretion in turkeys only at high doses[5], and similar results were obtained here. In contrast, pure natural porcine VIP stimulated pancreatic secretion at relatively low doses. Figure 1 shows that both peptides evoked dose-related increases in the rate of flow of pancreatic juice, but only weakly stimulated the rate of pancreatic protein secretion. From these results it was calculated that on a molar basis VIP was 28.5 times more potent than porcine secretin in stimulating the flow of pancreatic juice (estimated from a comparison D_{50} values which were determined by a modification of the Dowd-Riggs transformation for dose-response data[6]). A similar although slightly lower value for the relative potency of the 2 peptides has also been obtained using a preparation of synthetic porcine VIP. Thus in conventional 2 + 2 assays the synthetic material was found to be 15–20 times more potent than porcine secretin in stimulating the flow of juice from the turkey pancreas.

In view of the fact that VIP is related in structure not only to secretin but also glucagon, it was thought of interest to test the action of the latter peptide on the avian pancreas. Results presented in the Table show that even in doses as high as 200 μg/kg a preparation of beef-pork glucagon (Sigma Chemicals) had no action on the flow of juice or protein from the resting pancreas in turkeys. The animals in which glucagon was tested did, however, respond to secretin and the C-terminal octapeptide of cholecystokinin (OP-CCK). The pancreatic responses to secretin were the same as those previously noted, while the responses to OP-CCK were characterized by increases in the rate of flow of juice accompanied by a large increase in the rate of protein secretion, and were

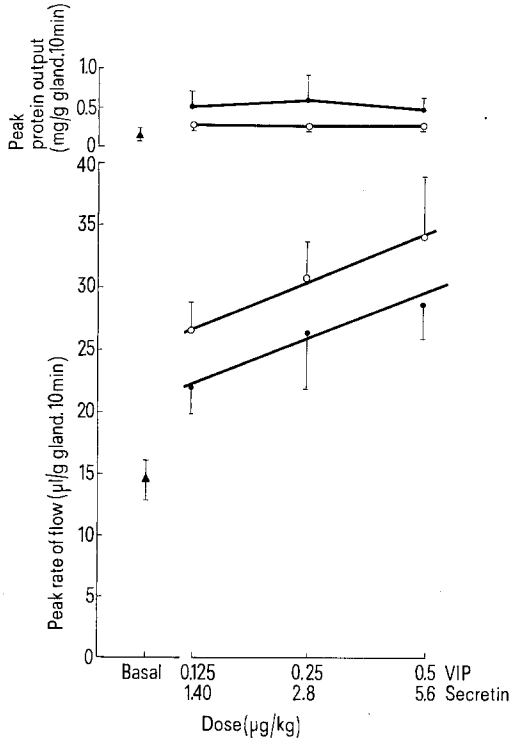

Fig. 1. The action of pure natural porcine secretin and pure natural porcine VIP on pancreatic secretion in 6 turkeys (mean body weight 2.5 kg). In calculating the dose of secretin it was assumed that 3.5 clinical units = 1 μg. The responses are expressed as the peak 10 min rate of secretion which occurred after the injections. The injections were made at 30 min intervals, and order of doses was determined on the basis of a 6×6 latin square. Each point represents the mean responses of 6 animals ± S.E.M. O, VIP; ●, Secretin.

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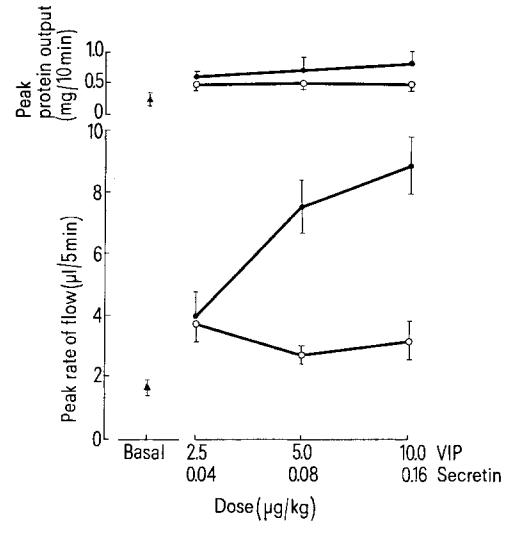

Fig. 2. The actions of synthetic porcine VIP and natural porcine secretin on pancreatic secretion in 5 rats (mean body weight 175 g). Responses are expressed as the peak 5 min rate of flow and peak 10 min rate of protein secretion from the pancreas. Injections were made at 30 intervals and order of doses randomized. Symbols as Figure 1.

therefore similar to the responses stimulated by OP-CCK and related peptides in the rat[7] and dog[8].

The actions of VIP and secretin on pancreatic secretion in the rat are shown in Figure 2. It is apparent that porcine secretin evoked dose-related increases in the flow of pancreatic juice at doses which were approximately 50 times less than those active in turkeys. In contrast VIP, in doses 20 times greater than those administered to the turkey, evoked only small increases in the flow of juice, although these were significantly greater than do basal levels ($P < 0.05$). However it is obvious from Figure 2 that the responses to VIP in the rat were not dose-related so that a precise estimate of the relative potency of secretin and VIP in this species cannot be made.

The action of peptides on the rate of flow and protein secretion from the turkey pancreas

Peptide	Rate of flow (µl/g gland. 10 min)	Rate of protein secretion (mg/g gland. 10 min)
Basal secretion	12.1 ± 2.0	0.31 ± 0.02
Porcine secretin (5 µg/kg)	23.1 ± 5.8[a]	0.76 ± 0.25[a]
OP-CCK (0.5 µg/kg)	28.7 ± 7.2[a]	6.5 ± 1.6[a]
Porcine/bovine glucagon (200 µg/kg)	9.9 ± 1.8	0.30 ± 0.17

The peptides were administered i.v., in random order, to each of 5 turkeys. The response was taken as the rate of flow and protein secretion which occurred in the 10 min period immediately after the injections. At least 30 min was allowed to elapse between injections. Values are means ± S.E.M., [a]denotes values significantly different from basal ($p < 0.01$).

It is clear from these results that VIP was a strong stimulant of the flow of pancreatic juice in the turkey, but not in the rat or cat[2]. The strong action of VIP on the avian pancreas indicates that in one respect at least this peptide resembles avian secretin more closely than either porcine secretin or glucagon. Although there have been several discussions recently about the evolutionary significance of similarities in the aminoacid sequences of mammalian secretin, glucagon, VIP and various other peptides[9–11], relatively little is understood of the phylogenetic relationships between these molecules. The results presented here raise the possibility that VIP and avian secretin may have inherited from a common ancestor features which are not present in modern mammalian secretin and glucagon[12].

Zusammenfassung. Das vasodilatierende intestinale Peptid vom Schwein hat beim Truthahn, nicht aber bei der Ratte, eine starke pankreassekretionfördernde Wirkung. Somit besteht beim Vogelsekretin Verwandtschaft bezüglich Wirkung und unterscheidet sich dadurch vom Schweinesekretin oder vom Glucagon.

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Starvation and Gastrin Storage in the Pyloric Antral Mucosa of Male Rabbits

The second gastrointestinal hormone, gastrin, was discovered by Edkins[1] and it has since engaged the attention of many workers. So far, the natural form of forms in which gastrin exist in the pyloric mucosa and in circulation are unknown. The mode of synthesis, the factors controlling the rate of synthesis, and even the storage of gastrin in the pyloric mucosa are still not known. This work was done to determine the effect of starvation on gastrin storage and activity in the pyloric mucosa of male rabbits.

Materials and methods. Thirty-six 6 – $6^1/_2$-month-old male rabbits weighing between 2.3 – 2.5 kg were used for the experiments. 9 of the rabbits were taken out during feeding, weighed and then killed, while feeding was discontinued for the remaining 27 rabbits. Blood was collected from each of the killed rabbits and the blood glucose content was determined separately by the glucose oxidase method using *O*-dianisidine[2]. The stomachs and the livers of the rabbits were immediately removed. The livers were weighed, while the stomachs were cut open, washed clean with water, and the pyloric antrum cut out. The pyloric mucosa was scraped off carefully from the underlying musculature. The pyloric mucosae from the 9 stomachs were divided into 3 batches, each containing pyloric mucosae from 3 stomachs. These were then separately weighed and subjected to the gastrin extraction procedure described by Blair, Harper, Lake, Reed and Scratcherd[3].

12 h after last contact with food but with free supply of water, 9 other rabbits were weighed, killed and the blood, livers and stomachs treated as in the first group. 36 h after last contact with food but with free supply of water, 9 other rabbits were treated as in the first 2 groups. 72 h after last contact with food but also with free supply of water, the remaining 9 rabbits were weighed killed and treated as in the first 3 groups of rabbits. The extracts obtained from the 4 groups of male rabbits were dried and the quantity of powdered extract per gramme wet weight of the different pyloric mucosa was determined. Solutions of known concentrations of the extracts were prepared using normal saline (0.15 *M*) as solvent.

Assay of the mucosal extracts. The continuous stomach perfusion technique described by Ghosh and Schild[4] was modified for the assay. Male Wistar strain rats weighing between 180–210 g were used. The rats were anaesthetised with urethane in 25% w/v solution given i.p. at 0.6 ml/100 g body weight. The operative procedures on the rats were similar to those described by Ghosh and Schild[4] but the femoral vein was cannulated

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