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Human Pancreatic Secretory Trypsin Inhibitor

Distribution, Actions and Possible Role in Mucosal Integrity and Repair

Key Words

Cholecystokinin
Ulceration
Mucus
Mucosal integrity

Abstract

Pancreatic secretory trypsin inhibitor is a potent protease inhibitor which was originally identified in the pancreas. It has subsequently been shown to be present in mucus-secreting cells throughout the gastrointestinal tract and also in the kidney, lung and breast. Its major roles are likely to be to prevent premature activation of pancreatic proteases and to decrease the rate of mucus digestion by luminal proteases within the stomach and colon. In addition, PSTI increases the proliferation of a variety of cell lines and stimulates cell migration, possibly acting via the EGF receptor. These findings suggest that PSTI may also be involved in both the early and late phases of the healing response following injury. Further studies including the production of transgenic over-expression and knockout models should help elucidate the physiological function of this peptide.

Introduction

Historical Perspective

Protease inhibitors are ubiquitous throughout the plant and animal kingdom. The known inhibitors are not all homologous but consist of several families which have probably evolved by convergent evolution [1].

The first identification of an enzyme inhibitor within the pancreas was made in 1936. It is known as the basic pancreatic trypsin inhibitor, the Kunitz inhibitor, or aprotinin, and is marketed by Bayer as 'Trasylol'. This inhibitor strongly inhibits trypsin, chymotrypsin, plasmin and kallikrein and is present in the mast cells of the pancreas and other organs of some ruminant species [2], but is not secreted in pancreatic juice. The human pancreas, however, does not contain an aprotinin (Kunitz)-type homologue.

The discovery of a second type of trypsin inhibitor in the pancreas was not made until 1948 when Kazal et al. [3] purified bovine pancreatic secretory trypsin inhibitor (PSTI) from a side fraction of a commercial insulin process. Bovine PSTI can be distinguished from the bovine Kunitz inhibitor on the basis of its lack of inhibitory effect on chymotrypsin and pancreatic kallikrein [4]. The sequence of human PSTI was identified in 1977 as being 56 amino acid residues long with strong sequence homology with bovine PSTI. Human PSTI inhibits potently the proteolytic activity of trypsin and other serum proteases [5].

In addition to being isolated from the human pancreas, other groups noted increased concentrations of a trypsin inhibitor in the urine of women with gynecological malignancies [6]. They named this peptide 'tumour-associated trypsin inhibitor' (TATI), it was subsequently realised that TATI and PSTI were the same molecule.

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	-5	1	10	20	30	40	50
Human		Asp-Ser-Leu-Gly-Arg-Glu-Ala-Lys	Cys-Tyr-Asn-Glu-Leu-Asn	Gly-Cys-Thr-Lys-Ile-Tyr-Asp-Pro-Val	Cys-Gly-Thr-Asp		
Rat I		Gly-Asn-Pro-Pro-Ala-Glu-Val-Asn-Gly-Lys-Thr-Pro-Asn	Cys-Pro-Lys-Gln-Ile-Met	Gly-Cys-Pro-Arg-Ile-Tyr-Asp-Pro-Val	Cys-Gly-Thr-Asn		
Rat II		Lys-Val-Ile-Gly-Lys-Lys-Ala-Lys	Cys-Pro-Asn-Thr-Leu-Val	Gly-Cys-Pro-Arg-Asp-Tyr-Asp-Pro-Val	Cys-Gly-Thr-Asp		
Porcine I		Thr-Ser-Pro-Gln-Arg-Glu-Ala-Thr	Cys-Thr-Ser-Glu-Val-Ser	Gly-Cys-Pro-Lys-Ile-Tyr-Asn-Pro-Val	Cys-Gly-Thr-Asp		
Bovine		Asn-Ile-Leu-Gly-Arg-Glu-Ala-Lys	Cys-Thr-Asn-Glu-Val-Asn	Gly-Cys-Pro-Arg-Ile-Tyr-Asn-Pro-Val	Cys-Gly-Thr-Asp		
Ovine		Asn-Ile-Leu-Gly-Arg-Glu-Ala-Lys	Cys-Thr-Asn-Glu-Val-Asn	Gly-Cys-Pro-Arg-Ile-Tyr-Asn-Pro-Val	Cys-Gly-Thr-Asp		
Canine		Asn-Asn-Met-Leu-Gln-Arg-Gln-Ala-Asn	Cys-Asn-Leu-Lys-Val-Asn	Gly-Cys-Asn-Lys-Ile-Tyr-Asn-Pro-Ile	Cys-Gly-Ser-Asp		
Human		Gly-Asn-Thr-Tyr-Pro-Asn-Glu-Cys-Val	Leu-Cys-Phe-Glu-Asn-Arg-Lys-Arg-Gln	Tyr-Ser-Ile-Leu-Ile-Gln-Lys-Ser	Gly-Pro-Cys		
Rat I		Gly-Ile-Thr-Tyr-Pro-Ser-Glu-Cys-Ser	Leu-Cys-Phe-Glu-Asn-Arg-Lys-Phe-Gly	Thr-Ser-Ile-His-Ile-Gln-Arg-Arg-Gly	Thr-Cys		
Rat II		Gly-Lys-Thr-Tyr-Ala-Asn-Glu-Cys-Ile	Leu-Cys-Phe-Glu-Asn-Arg-Lys-Phe-Gly	Thr-Ser-Ile-Arg-Ile-Gln-Arg-Arg-Gly	Leu-Cys		
Porcine I		Gly-Ile-Thr-Tyr-Ser-Asn-Glu-Cys-Val	Leu-Cys-Ser-Glu-Asn-Lys-Arg-Gln	Thr-Pro-Val-Leu-Ile-Gln-Lys-Ser	Gly-Pro-Cys		
Bovine		Gly-Val-Thr-Tyr-Ser-Asn-Glu-Cys-Leu	Leu-Cys-Met-Glu-Asn-Lys-Glu-Arg-Gln	Thr-Pro-Val-Leu-Ile-Gln-Lys-Ser	Gly-Pro-Cys		
Ovine		Gly-Val-Thr-Tyr-Ser-Asn-Glu-Cys-Leu	Leu-Cys-Met-Glu-Asn-Lys-Glu-Arg-Gln	Thr-Pro-Val-Leu-Ile-Gln-Lys-Ser	Gly-Pro-Cys		
Canine		Gly-Ile-Thr-Tyr-Ser-Asn-Glu-Cys-Leu	Leu-Cys-Leu-Glu-Asn-Lys-Arg-Gln	Thr-Ser-Ile-Leu-Val-Glu-Lys-Ser	Gly-Pro-Cys		

The entire human PSTI gene has been characterised, it is 7.5-kb long separated into 4 exons and is located on chromosome 5 [7]. The gene product consists of 79 amino-acid residues which includes a 23 amino acid signal peptide. This results in a mature peptide of 56 amino acids which has a molecular weight of about 6,500 with 3 intra-chain disulphide bridges in a Cys⁹-Cys³³, Cys¹⁶-Cys³⁵, and Cys²⁴-Cys⁵⁶ configuration [5]. As can be seen from figure 1, the sequence of PSTI is highly conserved across species with complete conservation of the six Cys residues involved in disulphide bridging. Interestingly, the reactive site of human PSTI, Lys¹⁸, which interacts with the serine proteases is substituted by another basic side chain amino acid, arginine, in some species (fig. 1).

Distribution of Human PSTI

Until recently, PSTI was thought to be present only in the pancreas where its sole role was thought to be to prevent premature activation of pancreatic proteases. However, PSTI has subsequently been identified in mucus producing cells throughout the gastrointestinal tract [8, 9] (fig. 2) and in a range of other tissues including lung, liver, kidney, ovary, breast and in the collecting tubules and transitional epithelium of the renal pelvis [10, 11]. Mucosal levels of PSTI in the normal gastrointestinal tract range from about 1,000 ng/mg protein in the gastric antrum to about 200 ng/mg protein in the colon [9]. PSTI is also present in plasma at a concentration of about 4–25 ng/ml under normal circumstances [12] and has a circulatory half-life of about 8 min, the major site of excretion being via the kidneys [13]. Following removal of the pancreas, normal serum PSTI levels are found, suggesting that the pancreas is not the major source of circulating human PSTI [14].

In situ hybridisation studies have confirmed this distribution of PSTI although PSTI mRNA was not found in the renal collecting tubules [11]. This is probably because PSTI is filtered in the kidney and taken up by the proximal tubules where it is degraded. The finding of PSTI

peptide in the normal breast [11] raises the possibility that it is secreted into breast milk, although quantitation in milk has not been performed. PSTI has also been shown to be secreted into gastric juice and is the only protease inhibitor known to be secreted into the intestinal lumen [15].

Possible Biological Functions of Human PSTI

Research into the role of human PSTI has concentrated on 3 main areas.

PSTI Working as a Cholecystokinin (CCK)-Releasing Peptide

Human PSTI has amino acid sequence homology with the two forms of PSTI present in the rat (fig. 1), one of which (PSTI-1) acts as a cholecystokinin releasing peptide [16]. There has therefore been great interest as to whether human PSTI plays a similar role.

Studies in rats showed that diversion of pancreatic juice away from the intestinal lumen results in increased pancreatic secretion. This increase in pancreatic secretion could be suppressed by adding trypsin or chymotrypsin to the intestinal lumen, leading to the conclusion that pancreatic secretion was under feedback regulation acting via luminal proteases [17]. In addition, it was suggested that the increase in pancreatic secretion which occurs following infusion of trypsin inhibitors into the duodenum of rats was due to the trypsin inhibitors interfering with the feedback loop [17]. This increase in pancreatic secretion was subsequently shown to be mediated via increases in plasma CCK levels.

Negative feedback of CCK release in the rat has subsequently been attributed to the presence of trypsin-sensitive CCK releasing peptides in pancreatic and intestinal juice. The CCK releasing peptide present in the pancreatic juice has been sequenced and shown to be rat PSTI-1, also called 'monitor peptide' [16].

Although the mechanisms regulating pancreatic secretion in man are probably more complicated than in the rat, a similar feedback control has been shown in humans; patients who have had pancreatic juice diverted away from the duodenum, so that the juice drains externally, have increased pancreatic secretion. This high output of pancreatic juice can be decreased by the addition of trypsin or pancreatic juice into the intestinal lumen [18]. A series of experiments have subsequently been performed to examine this idea. We showed that intra-luminal infusions of human PSTI into an isolated section of human

Fig. 1. The sequence of pancreatic secretory trypsin inhibitor (PSTI) is highly conserved across species. Amino acids identical in all 7 structures are indicated by shading and include all six Cys residues involved in the intrachain Cys-Cys disulphide bridging.

Fig. 2. Immunohistochemical staining for the presence of pancreatic secretory trypsin inhibitor (PSTI). Figures adapted from Playford et al. [24]. **a** Human pancreas showing PSTI is present in large amounts in the exocrine cells of the pancreas (cytoplasmic dark staining) but not in the endocrine cells (the islet of Langerhans being negative at the centre of the figure). **b** Normal human stomach showing large amounts of PSTI in the superficial (mucus producing) cells of the stomach. **c** Biopsy from a patient who has atrophic gastritis (and a gastric ulcer) showing markedly reduced tissue PSTI levels.

a

b

c

Fig. 3. Damage to gastric mucus as assessed by gel filtration. Figure adapted from Playford et al. [15]. 6 mg of normal human mucus glycoprotein derived from 5 ml of gastric juice was incubated at 37°C for 1 h with 100 µg of trypsin with and without 26 µg of human PSTI (giving equimolar amounts of PSTI and added trypsin). Samples were analysed using a 16 × 900 mm Sephacryl S-500 column. Eluates were analysed for mucus carbohydrate using the phenol/sulphuric acid method. This system separates molecules according to size, the largest eluting first. Normal mucus elutes as an early single peak indicated by an arrow in the void volume (V_0). The position of the column's total bed volume (V_t) is also shown. **a** After incubation with trypsin, the mucus elutes later indicating breakdown of its structure to smaller fragments. **b** The presence of PSTI in the incubation medium prevented this destruction of mucus.

duodenum *in vivo* did not cause an increase in pancreatic secretion or CCK release [19]. This suggests that human PSTI does not play an analogous role to rat monitor peptide. Our conclusion is supported by the finding that infusion of recombinant human PSTI into the rat small intestine does not stimulate pancreatic secretion [20]. The control of pancreatic secretion in humans therefore appears to be more complicated than in the rat and other control mechanisms, besides the presence or absence of proteases within the gut lumen, are likely to be at least as relevant.

PSTI, Working as a Protease Inhibitor, Protecting the Mucus Layer

The identification of PSTI in mucus-secreting cells throughout the gastrointestinal tract suggests that PSTI may play a vital role in the protection of mucus against excessive destruction by luminal proteases. Mucus is a key defensive component of the gut, forming a continuous visco-elastic layer which is adherent to the epithelium within the stomach and colon. The mucus layer within the stomach, in combination with secreted bicarbonate, acts as a diffusion barrier, allowing the epithelial surface to be maintained at a neutral pH despite the gastric juice having a pH of about 2. Mucus also acts as a lubricant, protecting the stomach from direct trauma due to ingested food particles and facilitating the passage of faeces in the colon. It also prevents direct contact between luminal bacteria and the colonic mucosa.

The thickness of the mucus gel in normal subjects is between 50 and 450 µm in unfixed specimens. In patients with gastric ulcer the mucus layer is highly abnormal microscopically showing 'roughening' of the mucus layer, which contains a mixture of mucus and cellular debris, but also consists of lower-molecular-weight forms [21]. Abnormalities of gastric mucus have also been described in patients suffering from chronic gastritis without an ulcer [22]. This mucus is likely to be structurally weaker than normal as the stability and strength of mucus, assessed by its ability to resist deforming stress, is reduced in mucus consisting of glycoproteins with a lower molecular weight [23]. We have shown that patients with gastritis have reduced PSTI levels [24], (fig. 2c) and that therapeutic doses of the prostaglandin E_2 analogue, misoprostal, stimulates a threefold increase in gastric secretion of PSTI [15]. Alteration in the protease/anti-protease balance within the mucus layer may therefore be relevant in explaining some of these abnormalities (fig. 3) and also influencing the healing process.

Abnormalities of the mucus layer are also seen in patients with inflammatory bowel disease. An imbalance

Fig. 4. Effect of human purified PSTI on wound healing as assessed by cell migration. Figure adapted from Marchbank et al. [11]. The addition of PSTI (1–10 $\mu\text{g/ml}$) to wounded monolayers of the human colonic cells line HT-29 stimulates a dose-dependent increase in the rate of migration of the cells. This effect could be blocked by adding a PSTI-neutralising or an EGF receptor-blocking antibody (not shown). The addition of EGF at a dose of 5 $\mu\text{g/ml}$ to the system (●) caused a similar increase in restitution as that seen when PSTI was added at a dose of 10 $\mu\text{g/ml}$ (△).

in the proteolytic/anti-proteolytic load on the mucus layer is probably important in explaining the increased turnover of mucus in these patients; luminal serine proteases are increased (probably of bacterial origin) [25] and colonic mucosal PSTI levels are concomitantly reduced [26]. Importantly, the reduction in mucosal PSTI levels remains even in quiescent colitis. This has led us to suggest that the reduced mucosal PSTI levels found in these patients represents a long term reduction in a mucosal defence mechanism, leading to an increased susceptibility to further episodes of injury [26].

PSTI, Working as a Growth Factor to Influence Growth and Repair

There are many structural similarities between PSTI and the potent growth factor epidermal growth factor (EGF); both have similar numbers of amino-acid residues (56 and 53, respectively), molecular weights (about 6 kD), three intra-chain disulphide bridges and limited sequence homology. There is 50% gene sequence homology between PSTI and EGF; however, the intron-exon junctions are not related suggesting convergent evolution [27].

In 1985, Ogawa et al. [28] reported that human PSTI was mitogenic for human fibroblasts. The idea that PSTI acts as a classical growth factor was supported by the findings that that PSTI is secreted by human hepatoma cells and stimulates growth of human endothelial cells [29]. Studies of the mechanisms of PSTI-induced growth on various cell lines, however, have not given consistent results. Stimulation of growth of endothelial cells can be induced by soya bean trypsin inhibitor, human α_1 -proteinase inhibitor and bovine PSTI in addition to human PSTI [29]. This suggests that the trypsin inhibitor activity is an important component in stimulating growth. However, these other trypsin inhibitors do not induce growth of human or mouse fibroblasts or the rat pancreatic carcinoma cell line AR4-2J cells suggesting an interaction of PSTI with a specific receptor [29, 30]. The identity of this receptor(s) is, however, still unclear with continuing controversy as to whether PSTI binds to the classical EGF receptor or its own distinct receptor.

Some studies support the concept that PSTI binds to the EGF receptor. Rat monitor peptide (PSTI-I) has been reported to compete with mouse EGF for binding to the receptors of Swiss 3T3 fibroblasts [31] and an EGF receptor-blocking antibody removed the promigratory effects of PSTI on human HT-29 cells [11]. In contrast, other studies suggest a distinct receptor. Binding of human [^{125}I]PSTI to Swiss 3T3 cells has been reported to be displaceable by cold PSTI but not by human EGF [32] and a similar result was seen using the rat pancreatic cancer cell line AR4-2J [30]. Additional stimulation of DNA synthesis of Swiss 3T3 fibroblasts is possible using EGF when stimulation by human PSTI is already at a maximum, and vice versa [32], also supporting the concept of a distinct PSTI receptor.

PSTI in Mucosal Repair

When an acute injury occurs, the defect is rapidly covered by a mucoid cap consisting of mucus, fibrin and degenerative cells. Surviving cells from the edge of the wound rapidly migrate over the denuded area in a process called epithelial restitution. This process begins within the first hour following injury and is followed by an increase in proliferation 24–48 h later [33]. The physiological stimuli involved in epithelial restitution are unclear but we have recently shown that PSTI stimulates cell migration of the human colonic carcinoma cell line HT-29 cells in an in vitro wounding model of epithelial restitution (fig. 4). This effect could be inhibited by the addi-

tion of an EGF receptor-blocking antibody. PSTI may therefore be involved in both the early and late responses to intestinal injury. Additional evidence of the role of PSTI in mucosal repair is that PSTI is produced by the ulcer-associated cell lineage (UACL) [26]. This novel glandular structure is produced at sites of damage in conditions such as peptic ulceration and inflammatory bowel disease. The UACL secretes several peptides thought to be involved in mucosal repair, such as epidermal growth factor and the trefoil peptides in addition to PSTI onto the ulcerated surface [34].

PSTI may therefore play key roles in maintaining epithelial integrity and stimulating both the early and late response to injury. During the initial phase of repair, PSTI may stabilise the overlying 'mucoic cap' from digestion and stimulate surviving cells at the edge of the wound to migrate over the denuded area. During the later phase of repair, PSTI may also be directly involved in the increase in proliferation.

Conclusions

Although it is now 20 years since human PSTI was sequenced, there is a resurgence in interest in its physiological role. It seems unlikely that it is important in CCK physiology but may well be a key player in protection of the mucus layer from excessive digestion and in stimulating repair at site of injury. Studies utilising transgenic overexpression and knockout models should help elucidate its physiological role further.

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References

- 1 Bode W, Huber R: Natural protein proteinase inhibitors and their interaction with proteinases. *Eur J Biochem* 1992;204:433-451.
- 2 Fritz H, Kruck J, Russe I, Liebich HG: Immunofluorescence studies indicate that the basic trypsin-kallikrein-inhibitor of bovine organs (Trasylol) originates from mast cells. *Hoppe Seylers Z Physiol Chem* 1979;360:437-444.
- 3 Kazal LA, Spicer DS, Brahinsky RA: Isolation of a crystalline trypsin inhibitor-anticoagulant protein from the pancreas. *J Am Chem Soc* 1948;70:304-340.
- 4 Laskowski M, Wu FC: Temporary inhibition of trypsin. *J Biol Chem*. 1953;204:797-805.
- 5 Bartelt DC, Shapanka R, Green LJ: The primary structure of the human pancreatic secretory trypsin inhibitor. *Arch Biochem Biophys* 1977; 179:189-199.
- 6 Huhtala ML, Kahanpaa K, Seppala M, Halila H, Stenman UH: Excretion of a tumour associated trypsin inhibitor (TATI) in urine of patients with gynaecological malignancy. *Int J Cancer* 1983;31:711-714.
- 7 Horii A, Kobayashi T, Tomita N, Yamamoto T, Fukushima S, Murotsu T, Ogawa M, Mori T, Matsubara K: Primary structure of human secretory trypsin inhibitor (PSTI) gene. *Biochem Biophys Res Commun* 1987;149:635-641.
- 8 Shibata T, Ogawa M, Matsuda K, Miyauchi K, Yamamoto T, Mori T: Purification and characterisation of pancreatic secretory trypsin inhibitor in human gastric mucosa. *Clin Chim Acta* 1986;159:27-36.
- 9 Freeman TC, Playford RJ, Quinn K, Beardshall K, Calam J: Pancreatic secretory trypsin inhibitor in gastrointestinal mucosa and gastric juice. *Gut* 1990;31:1318-1323.
- 10 Shibata T, Ogawa M, Takata N: Distribution of pancreatic secretory trypsin inhibitor in various human tissues and its inactivation in the gastric mucosa. *Res Commun Pathol Pharmacol* 1987;55:243-248.
- 11 Marchbank T, Chinery R, Hanby AM, Poulson R, Elia G, Playford RJ: Distribution and expression of pancreatic secretory trypsin inhibitor and its possible role in epithelial restitution. *Am J Pathol* 1996;148:715-722.
- 12 Pezzilli R, Billi P, Plate L, Barakat B, Bongiovanni F, Miglioli M: Human pancreatic secretory inhibitor in the assessment of the severity of acute pancreatitis. *J Clin Gastroenterol* 1994;19:112-117.
- 13 Marks WH, Ohlsson K: Elimination of pancreatic secretory trypsin inhibitor from the circulation. *Scand J Gastroenterol* 1983;18:955-959.
- 14 Halila H, Huhtala ML, Schroder T, Kiviluoto T, Stenman UH: Pancreatic secretory trypsin inhibitor-like immunoreactivity in pancreatized patients. *Clin Chim Acta* 1985;153: 209-216.
- 15 Playford RJ, Batten JJ, Freeman TC, Beardshall K, Vesey D, Fenn GC, Baron JH, Calam J: Gastric output of pancreatic secretory trypsin inhibitor is increased by misoprostol. *Gut* 1991;32:1396-1400.
- 16 Fukuoka SI, Scheele G: Complementary nucleotide sequence for monitor peptide, a novel cholecystokinin-releasing peptide in the rat. *Nucl Acids Res* 1989;17:10111.
- 17 Owyang C, May D, Louie DS: Trypsin suppression of pancreatic enzyme secretion. *Gastroenterology* 1986;91:637-643.
- 18 Owyang C, Louie DS, Tatum D: Feedback regulation of pancreatic enzyme secretion. *J Clin Invest* 1986;77:2042-2047.
- 19 Playford RJ, Freeman TC, Calam J: Is human pancreatic secretory trypsin inhibitor a CCK-releasing peptide in man? *Regul Pept* 1992;40: 228.
- 20 Miyasaka K, Guan DF, Liddle RA, Green GM: Feedback regulation by trypsin: Evidence for intraluminal CCK-releasing peptide. *Am J Physiol* 1989;257:G175-G181.
- 21 Allen A, Hutton DA, Leonard AJ, Pearson JP, Sellers LA: The role of mucus in the protection of the gastroduodenal mucosa. *Scand J Gastroenterol* 1986;(suppl 125):71-78.
- 22 Molinari F, Parodi MC, De Angelis P, Cheli R: Gastric mucus secretion in chronic gastritis. *Scand J Gastroenterol* 1984;19(suppl 92):167-171.
- 23 Bell AE, Sellers A, Allen A, Cuncliffe WJ, Morris ER, Ross-Murphy SB: Properties of gastric and duodenal mucus: Effect of proteolysis, disulphide reduction, bile, acid, ethanol, and hypertonicity on mucus gel structure. *Gastroenterology* 1985;88:269-280.

- 24 Playford RJ, Hanby AM, Quinn C, Calam J: Influence of inflammation and atrophy on pancreatic secretory trypsin inhibitor levels within the gastric mucosa. *Gastroenterology* 1994; 106:735–741.
- 25 Senda S, Fujiyama Y, Bamba T, Hosada S: Treatment of ulcerative colitis with camostat mesilate, a serine protease inhibitor. *Intern Med* 1993;32:350–354.
- 26 Playford RJ, Hanby AM, Patel K, Calam J: Influence of inflammatory bowel disease on the distribution and concentration of pancreatic secretory trypsin inhibitor within the colon. *Am J Path* 1995;146:310–316.
- 27 Scheving LA: Primary amino acid sequence similarity between human epidermal growth factor-urogastrone, human pancreatic secretory trypsin inhibitor and members of the secretin family. *Arch Biochem Biophys* 1983;226: 411–413.
- 28 Ogawa M, Tsushima T, Ohba Y: Stimulation of DNA synthesis in human fibroblasts by human pancreatic secretory trypsin inhibitor. *Res Commun Chem Pathol Pharmacol* 1985;50: 155–158.
- 29 McKeenan WL, Sakagami Y, Hoshi H: Two apparent human endothelial cell growth factors from hepatoma cells are tumor-associated proteinase inhibitors. *J Biol Chem* 1986;261: 5378–5383.
- 30 Freeman TC, Curry BJ, Calam J, Woodburn J: Pancreatic secretory trypsin inhibitor stimulates the growth of pancreatic tumour cells. *Gastroenterology* 1990;99:1414–1420.
- 31 Fukuoka SI, Fushiki T, Kitagawa Y, Sugimoto EI: Competition of a growth stimulating-/cholecystokinin (CCK) releasing peptide (monitor peptide) with epidermal growth factor binding to 3T3 fibroblasts. *Biochem Biophys Res Commun* 1987;145:646–650.
- 32 Niinobu T, Murata A, Nishijima J, Mori T: Identification and characterisation of receptors specific for human pancreatic secretory trypsin inhibitor. *J Exp Med* 1990;172:1133–1142.
- 33 Silen W: Gastric mucosal defence and repair; in Johnson LR (ed): *Physiology of the Gastrointestinal Tract*, ed 2. New York, Raven, 1987, pp 1044–1069.
- 34 Poulsom R, Wright NA: Trefoil peptides: A newly recognised family of epithelial mucin-associated molecules. *Am J Physiol* 1993;265: G205–213.

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