LETTERS TO THE EDITOR

Do trypsin 2- α -1-antitrypsin complexes occur naturally in the circulation?

EDITOR.—Some recent studies on the measurement of serum trypsin 2- α -1-antitrypsin complexes in the diagnosis of acute pancreatitis have piqued our interest,^{1,2} including that of Kempainen *et al* (*Gut* 1997;41:690-5). We published a couple of articles on trypsin 1- α -1-antitrypsin complexes in serum some years ago. Principally, we arrived at the same conclusion as the Finnish group—namely, that concentrations of these complexes in serum correlate with disease severity.³⁻⁵

We had some problems with our assay in that we very often found low but measurable concentrations (5-25 μ g/l) of trypsin 1- α -1-antitrypsin complexes in serum samples obtained from patients with no signs of acute pancreatitis. Our method was not sensitive enough to allow detailed characterisation of this low immunoreactivity and we were therefore not sure what it was comprised of. We did not publish these results as we found them hard to believe. If they were true it would mean that a substantial amount of trypsinogen was normally activated in and around the pancreas during the normal turnover of pancreatic enzymes (see later). Our hypothesis has always been that activation of trypsinogen to trypsin is a unique process that only occurs in acute pancreatitis or in the duodenum during normal digestion.⁶ Interestingly, Stenman's group have reported up to 32 μ g/l of the trypsin 2- α -1-antitrypsin complex in serum of patients without acute pancreatitis.^{1,2} The Finnish method should be sensitive enough to characterise this immunoreactivity and we therefore pose the following questions: Is this normal trypsin 2- α -1-antitrypsin-like immunoreactivity really a complex of trypsin 2 and α -1-antitrypsin? Is activated trypsin normally present in the systemic circulation? (We think that the latter is an important question particularly with regard to the pathophysiology of acute pancreatitis.) Is trypsinogen secreted basolaterally from the pancreas normally or does this occur only in acute pancreatitis?

One must also consider that only about 10% of activated trypsin is bound to α -1-antitrypsin in serum. Most trypsin (90%) is complexed to α -2-macroglobulin⁷ which means that the presence of trypsin 2- α -1-antitrypsin complexes at a concentration in the range of 20 μ g/l reflects the activation of a substantial amount of trypsinogen. Alternatively, the trypsin 2- α -1-antitrypsin complexes present in the circulation could result from reabsorption of trypsin from the intestine, a matter much disputed in the literature.

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Reply

EDITOR.—We thank Professor Ohlsson and Dr Borgström for their stimulating thoughts about the possible mechanisms that could explain the relatively high concentrations of trypsin complexes occurring in the circulation of apparently healthy subjects. They suggest that in healthy subjects trypsin activation occurs only in the gut and that the occurrence of activated trypsin in the circulation indicates absorption of active trypsin from the gastrointestinal tract. This notion may well be correct, but there are also other possible explanations. The presence of pancreatic enzymes in the circulation has also been explained on the basis of secretion of these enzymes through the basolateral membranes into the extracellular space by the so-called constitutive-like pathway.¹ This does not explain why trypsinogen is activated and complexed in the absence of pancreatitis. The fact that trypsinogen activation peptide is not detected in urine in the absence of pancreatitis² suggests that trypsinogen is not activated, but this may also be explained by low concentrations and insufficient sensitivity of the assay. Because our assay methods for trypsinogen and trypsin complexes are both sensitive and specific, we believe that trypsin complexes actually occur in circulation of healthy subjects. This could be explained by several mechanisms.

Trypsinogen leaking out of the pancreas may be activated by other proteases in the extracellular fluid or by autoactivation.³ Active trypsin complexes with plasma derived protease inhibitors in this compartment. Because the 58 kDa α -1-antitrypsin (AAT) diffuses more readily into extracellular space than the high molecular weight α -2-macroglobulin, most of the active trypsin leaking out may complex with AAT. Complex formation between trypsin and pancreatic secretory trypsin inhibitor (PSTI) is also likely to occur in the extracellular fluid of the pancreas, but so far we have not been able to demonstrate the trypsin-PSTI complex in the circulation. This may be due to further complexation with α -2-macroglobulin and rapid removal of this complex.

As Borgström and Ohlsson point out, the proportion of complexed trypsin is surprisingly high. However, the concentrations of free and complexed trypsins in the circulation do not need to correspond to those of activated trypsin and trypsinogen, respectively, escaping

from the pancreas. The plasma concentrations are dependent on both the rate of formation and disappearance. Based on the half-times of another protease-serpin complex, prostate specific antigen (PSA) in complex with α -1-antichymotrypsin and free PSA, 2-3 days and 1-2 hours,⁴ respectively, trypsin-AAT is probably eliminated much more slowly than trypsinogen. If this is the case, the proportion of activated trypsin escaping into the circulation may actually be very low although the proportion of total trypsin immunoreactivity in plasma is fairly high.

We have previously shown that trypsin immunoreactivity occurs in serum of patients without a pancreas.⁵ This trypsin may leak into the circulation from normal extrapancreatic tissues. Trypsinogen immunoreactivity has been shown to occur in Paneth cells of the small intestine and in the epithelium of bile ducts and peribiliary glands.^{6,8} We have also observed raised concentrations of trypsin 2-AAT complex in serum of patients with malignant diseases of the digestive tract.⁹ Thus, trypsinogen and trypsin inhibitor complexes in serum may also be derived from extrapancreatic tissues.

So far, these mechanisms are hypothetical and need to be verified experimentally.

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BOOK REVIEW

Bile Acids in Hepatobiliary Disease. Basic Research/Clinical Applications.

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