

## **Section 12**

### **LECTURE**

### **Physiology and Biochemistry of the Pancreas**

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### I. Ultrastructure of the Exocrine Pancreas

1. Acinar Unit:
  - Exocrine Cells: secrete zymogens and enzymes
  - Centroacinar and Intralobular Duct Cells: secrete bicarbonate and water. Utilize CFTR (chloride channel which activates the chloride-bicarbonate exchanger. Water is thought to move passively. These duct cells are the progenitor stem cells which give rise to duct as well as acinar cells. It is for this reason that most adenocarcinomas of the pancreas are of ductal origin. EGF as well as TGF receptors are found on the apical cell membrane of the duct cell, with increased numbers of receptors correlating with a more aggressive cancer.
2. Interlobular and Main Duct cells:
  - secrete mucin and bicarbonate (non CFTR pathway in the rodent).
3. Result: digestive enzymes secreted into a dilute alkaline fluid, optimal for enzyme activation in the duodenum.

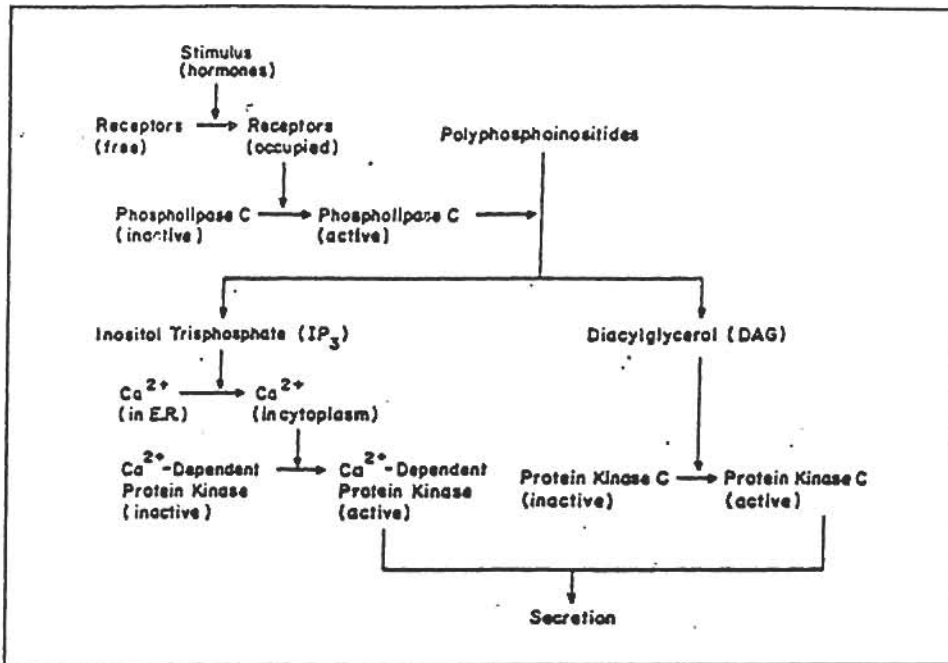
### II. The Acinar Cell:

The paradigm for studying basic cell function. This cell is specifically geared for large scale production and secretion of digestive enzymes.

1. The regulated pathway was determined in this cell: rough ER -> cis Golgi -> trans Golgi network -> Condensing Vacuole -> Zymogen Granule -> Apical plasmalemma. Involves the exocytosis of digestive enzymes.
2. The constitutive pathway is utilized in the production of constitutive proteins for the cell such as lysosomal enzymes.
3. A constitutive-like pathway has also been described whereby proteins which target to the lysosomal compartment for example, ultimately are secreted.

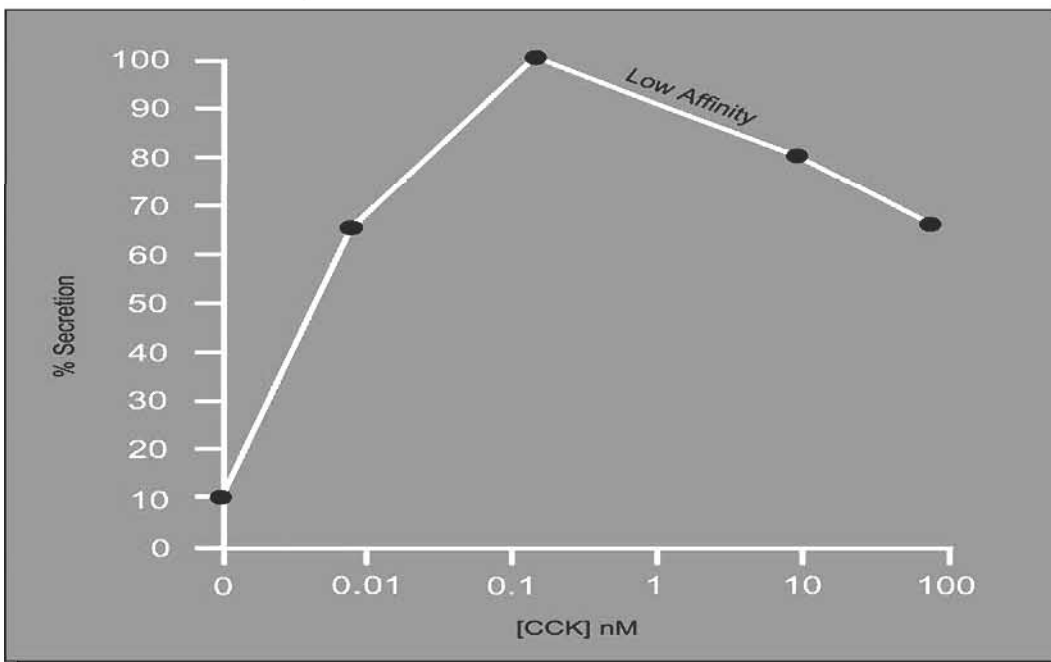
### III. Hormonal Regulation:

1. CCK: A and B receptors have been identified. These have been localized to the acinar (A) cell and brain (B) although both receptors are seen in various organs. Results in acinar cell secretion, gallbladder contraction, slows gastric emptying, and activates the satiety center. Acts via calcium and cGMP as 2nd messengers utilizing calcium calmodulin and PK-C kinase systems. Calcium is released from intracellular stores by the release of IP3 as shown below:



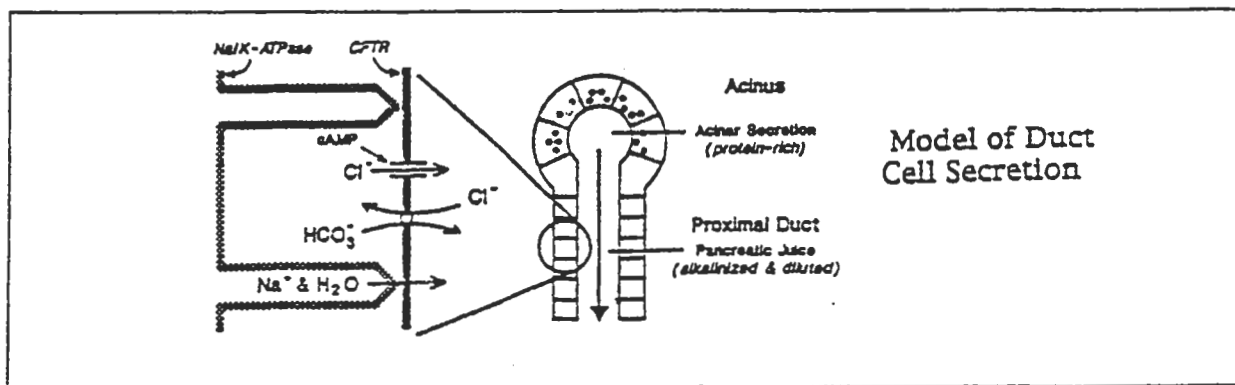
from Steer and Meldolesi NEJM 1987 pg 146: Stimulus-Secretion Coupling

2. The CCK receptor is regulated through 3 different mechanisms.
  - A. Desensitization through phosphorylation as a result of CCK occupation
  - B. short term downregulation via internalization into calveolae
  - C. long term downregulation via decreased synthesis and insertion
  
3. High and low affinity CCK A receptors: identified with different peptides. Results in biphasic secretion curve. Shown below is a typical dose response curve for secretion from acini (% secretion on Y axis) verses dose of CCK (nM) on X axis.



Of note, selective occupation of low affinity CCK receptors results in acute pancreatitis in animal models. This pathway does not utilize IP3 to increase intracellular calcium.

4. Acetylcholine:
  - A. stimulates acinar cell secretion.
  - B. CCK appears to stimulate acinar cell secretion, not directly under physiologic conditions, but rather via intrapancreatic cholinergic neurons.
  - C. may fine tune the response of the pancreas to a given stimulus.
  - D. acts through calcium as the 2nd messenger.
  
5. Secretin:
  - A. stimulates ductal bicarbonate secretion via CFTR.
  - B. uses cAMP as the 2nd messenger.
  
6. Somatostatin:
  - A. inhibits CCK and secretin release.
  - B. Inhibits intrapancreatic peptidergic neurons (effect blocked with tetrodotoxin)
  - C. may directly bind to receptors on the acinar cell to inhibit function (only shown in one animal model system).
  
7. Other hormones:
  - A. bombesin: hormone from frog skin. Acts via calcium signaling pathway.
  - B. VIP: similar action as with secretin.
  - C. Gastrin releasing peptide; neuropeptide Y, YY; calcitonin gene related peptide; galanin; substance P.



Taken from reference 9: CR Marino and FS Gorelick. Gastroenterology (1992) 103:681-693.

#### IV. Secretory Products:

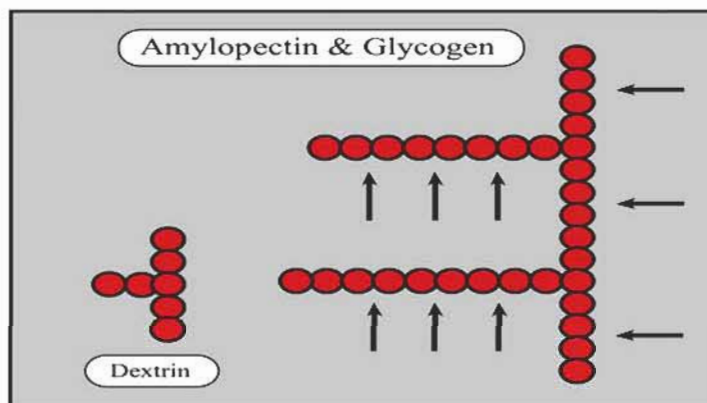
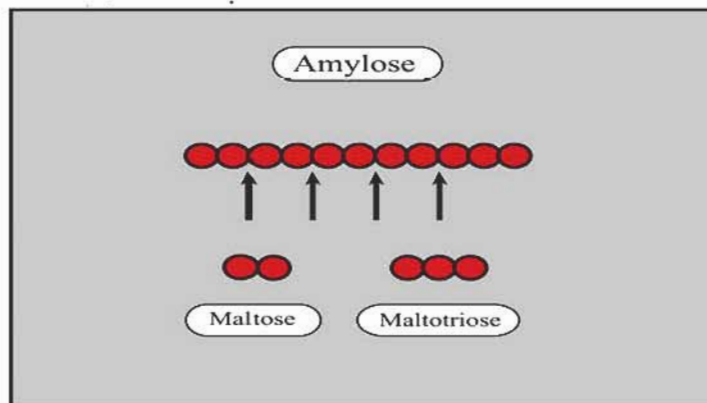
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|--|--|
| <p>A. <u>Proenzymes (Zymogens):</u><br/>         -trypsin(ogen):<br/>         -chymotrypsin(ogen):<br/>         -(pro)elastase:<br/>         -Exopeptidases:</p> | <p><u>bonds cleaved</u><br/>         arg, lys; ester and amide bonds<br/>         aromatic aa's (tyr, phe, tryp)<br/>         adjacent aliphatic aa's.<br/>         e.g. Carboxypeptidase A, B</p> |
|--|--|

B. These enzymes are produced as different isoelectric variants, e.g. anionic and cationic trypsinogen. This gives the organism a survival advantage in being able to digest various substrates under all conditions.

C. Amylase:

-hydrolyzes 1,4- $\alpha$ -glucose linkages of starch (amylose and amylopectin) and glycogen (similar to amylopectin but 1,6 branch points occur more frequently). Cannot break terminal 1,4 or 1,6 links.

-Enzymatic cleavage produces maltose (2 glucose units) and maltotriose (3 glucose units) from amylose. Alpha dextrins (4-8 glucose units) are additionally produced from amylopectin and glycogen. In the figure below, each circle represents a glucose molecule.



D. DNase, RNase, etc.

V. **Safe Secretion:**

Several safeguards in place:

- A. Enzymes and zymogens are synthesized as inactive precursors. Pancreatic juice is therefore devoid of proteolytic activity until it reaches the duodenum.
- B. Enzymes are synthesized and sorted into membrane-bound compartments within the acinar cell.
- C. Activation requires a trigger enzyme (enterokinase) outside the pancreas.

- D. Inhibitors present in the pancreas can trap up to 20% of potentially available trypsin activity.
- E. Intrapancreatic trypsin activates an enzyme(s) to degrade trypsinogen and other zymogens to inert products (mesotrypsin). Trypsin inhibitors include PSTI, mesotrypsin, and plasma protease inhibitors such as alpha-1 antitrypsin, alpha-2 macroglobulin, inter alpha-1 trypsin inhibitor, and alpha-1 antichymotrypsin.

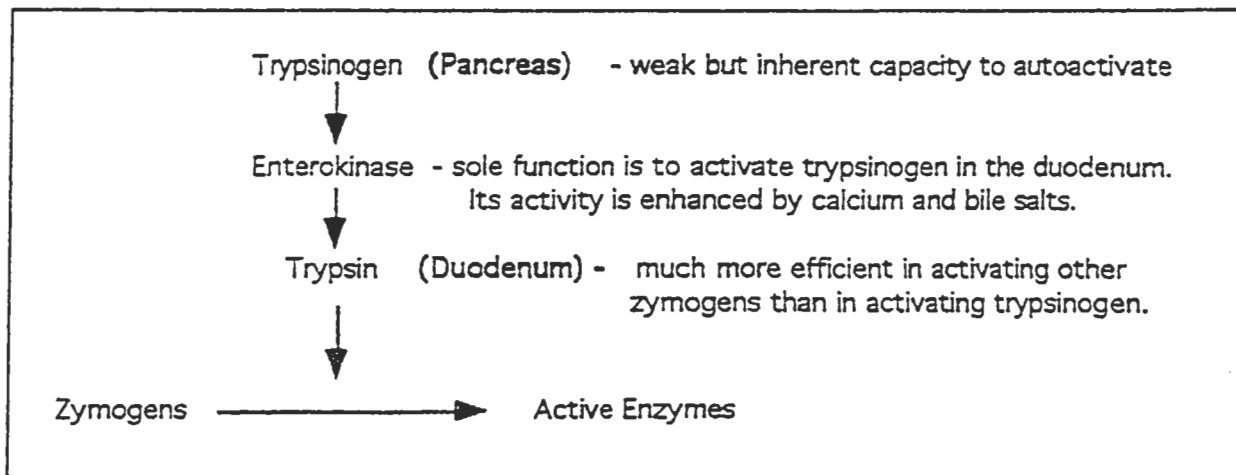
**VI. Integrated Response With the Duct Cell:**

1. It was previously thought that the only role of ductal bicarbonate secretion was to neutralize acid in the duodenum.
2. Recent data indicates that ductal bicarbonate secretion plays a primary role within the acinar lumen to:
  - A. Solubilize secretory proteins secreted into the acinar lumen.
  - B. Regulate membrane trafficking at the apical pole of the acinar cell.
  - C. This explains why CFTR is localized to the centroacinar and proximal intralobular duct cells adjacent to the acinar cells.

**VII. Regulation of Exocrine Pancreatic Secretion in Response to a Meal:**

The pancreas is a retroperitoneal organ which is comprised of endocrine and exocrine elements. It is the ultimate protein exporter, secreting up to 10 gms of protein per day.

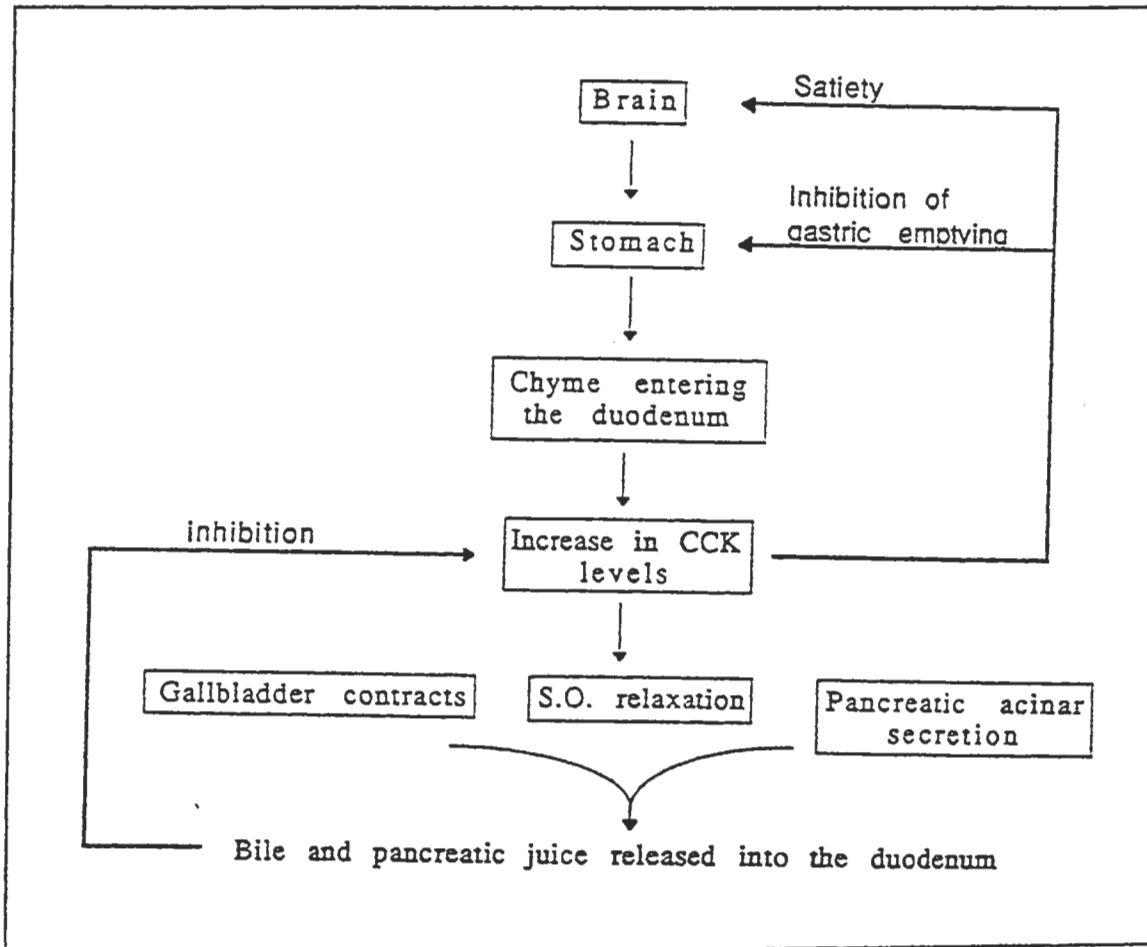
- A. Trypsinogen, converted to active trypsin in the duodenum by enterokinase, is the "trigger" enzyme which subsequently converts the other zymogens to active enzymes:



It is the premature activation of these digestive enzymes within the pancreatic gland which leads to pancreatitis.

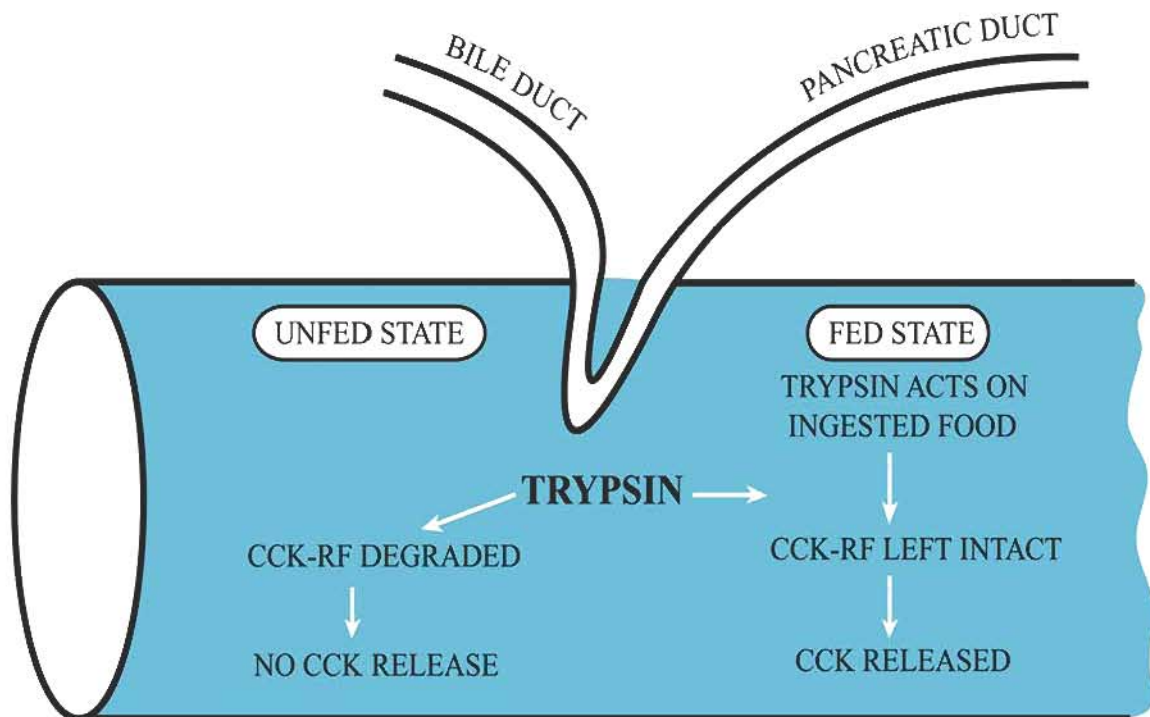
B. Physiologic Regulation of Pancreatic Secretion

1. Neurohormonal Control: Three phases have been described - cephalic, gastric, and intestinal. An important concept is that many overlying and even redundant regulated stimulatory mechanisms exist for the exocrine pancreas.
  - a. Cephalic Phase: low volume, protein rich juice. Can elicit up to 50% of the maximum pancreatic response. Is vagally mediated.
  - b. Gastric Phase: Production of small peptides and amino acids which are more amenable to digestion by the pancreatic enzymes. Most likely, the most important role of the stomach is to act as a reservoir and deliver small amounts of chyme to the duodenum for efficient pancreatic digestion.
  - c. Intestinal Phase: Occurs when chyme enters the duodenum. Quantitatively is the most important response. Acts on a pancreas already prestimulated by the cephalic and gastric phases. The magnitude of the response is determined by the length of time stimulants are exposed to the duodenum.



Regulation of GI function by CCK

2. Hormones:  
CCK: Distributed in CNS as well as GI tract. In response to chyme entering the duodenum, CCK levels rise. This stimulates smooth muscle of the gallbladder to contract, acinar cells to secrete digestive enzymes and the sphincter of Oddi to relax. CCK also inhibits further gastric emptying. CCK then acts on the satiety center in the brain.
3. Feedback Regulation of the Exocrine Pancreas:



Regulation of CCK release

- A. CCK releasing factor - trypsin sensitive factor produced in the duodenum which causes CCK to be released into blood. The mechanism of action of this feedback loop is as follows: ingestion of food ties up active trypsin allowing CCK factor to increase. Elevation of CCK (both locally and in the plasma) stimulates the pancreatic acinar cell to secrete. In the absence of substrate, trypsin secreted in the pancreatic juice digests CCK releasing factor resulting in low levels of CCK released into the bloodstream.
- B. Monitor peptide: 6 kD protein secreted into pancreatic juice from rats. Is also trypsin sensitive and a potent stimulus for CCK release. Similar mechanism as for CCK releasing factor. It "monitors" the duodenum for food. Unclear if this exists in humans.

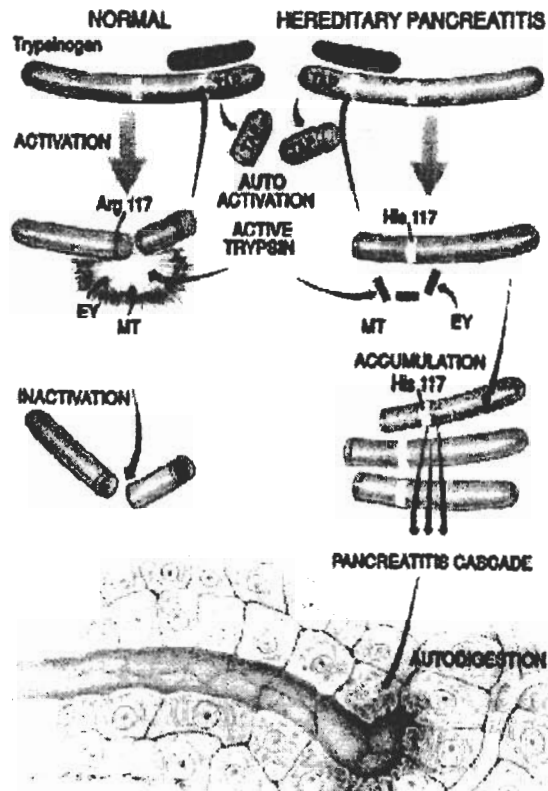


## Genetic

Preliminary data indicate that mutations of the Cystic Fibrosis gene as well as the Trypsin Inhibitor gene (SPINK-1) may explain the etiology in a number of patients previously labeled as idiopathic chronic pancreatitis.

## Hereditary Pancreatitis

Whitcomb et al (Nature Genetics 1996; 14:141-5) has recently discovered the gene responsible for this autosomal dominant disorder (see figure reproduced below from this article). Prematurely activated trypsin is normally inactivated by pancreatic secretory trypsin inhibitor (PSTI) within the zymogen granules or pancreatic duct system. In addition, prematurely activated trypsin degrades itself by cleavage at Arg 117. In Hereditary Pancreatitis, the cationic trypsinogen gene is mutated converting Arg 117 to His. This prevents autodigestion resulting in increased levels of active trypsin. Eventually the accumulated trypsin overwhelms PSTI resulting in pancreatitis.



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