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Duodenal Inflammation and Mucosal Integrity in Chronic Pancreatitis

Yoyo Zhang, MD, PGY-5

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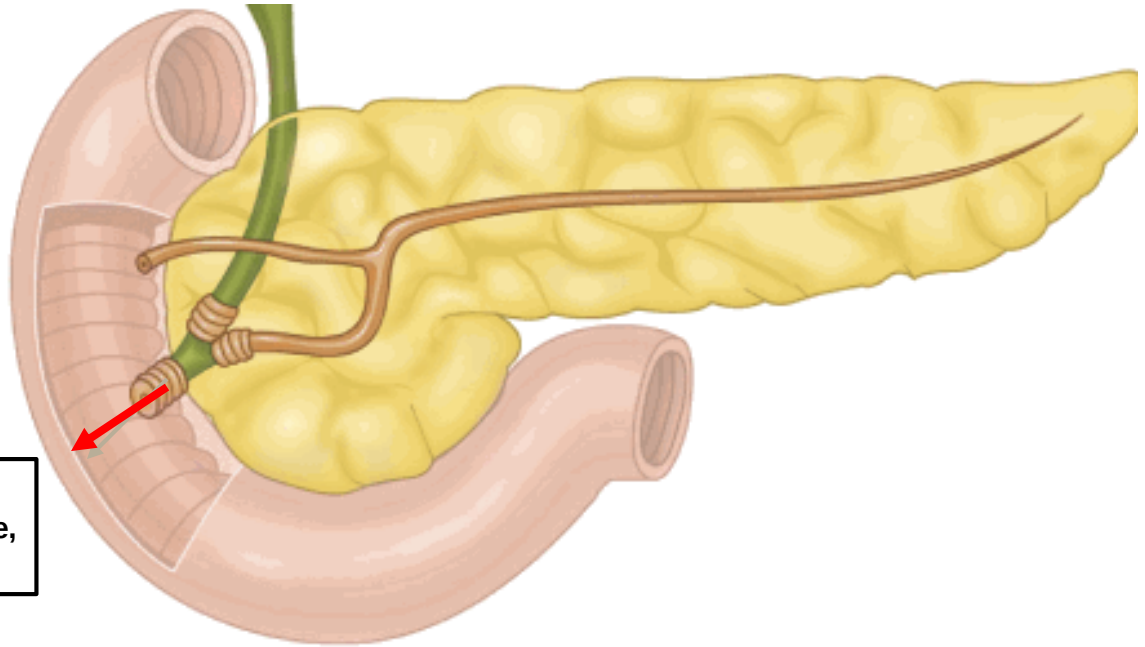
Duodenum - Pancreas



Release of digestive enzymes, bicarbonate, and anti-microbials

Recurrent Pancreatitis

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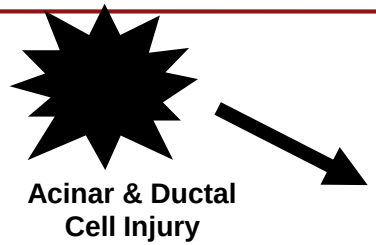
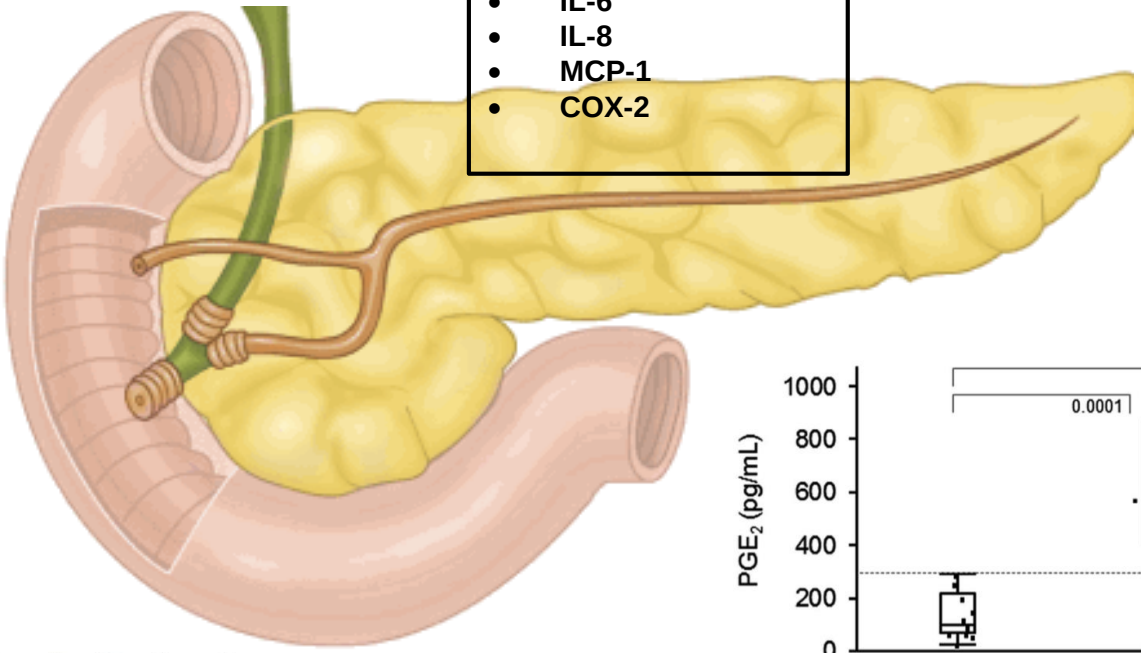
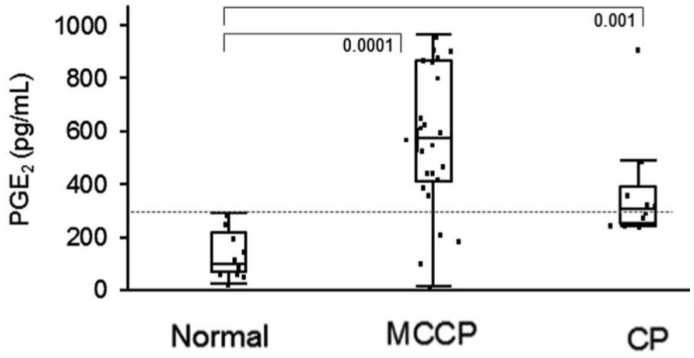


Acinar & Ductal Cell Injury

Release of pro-inflammatory molecules

- **PGE2**
- **TNFα**
- **IL-1**
- **IL-6**
- **IL-8**
- **MCP-1**
- **COX-2**





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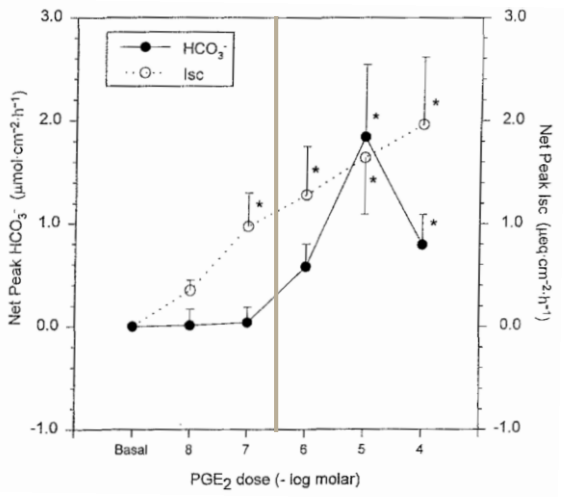
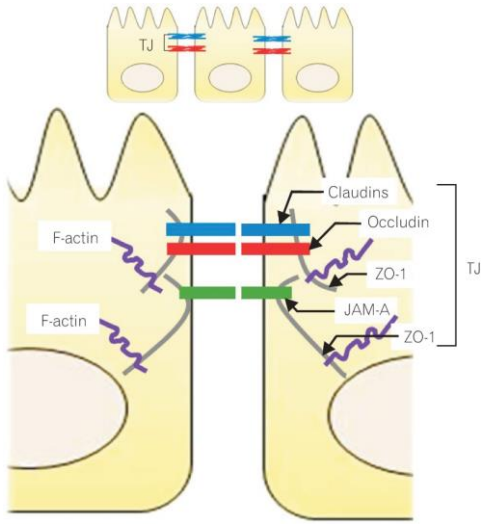
Clin Transl Gastroenterol. 2015 Jan 29;6:e72

PGE_2 in the Duodenum

- Gastric acid stimulates PGE_2 release
- PGE_2 dose-dependently increases duodenal bicarbonate secretion
- Unclear what effect chronic PGE_2 exposure has on the intestine
  - Inflammation? Alteration in immune cell activation.
  - Down-regulation of receptors?
  - Colon (Caco-2 cells): Prolonged exposure to PGE_2 increases intestinal epithelial permeability through redistribution of ZO-1, claudin-4, and occludin.
- Intestinal permeability and endotoxemia have been proposed as contributing to AP pathogenesis.
  - **It is unclear if intestinal permeability or intestinal function is impaired in CP pathogenesis.**



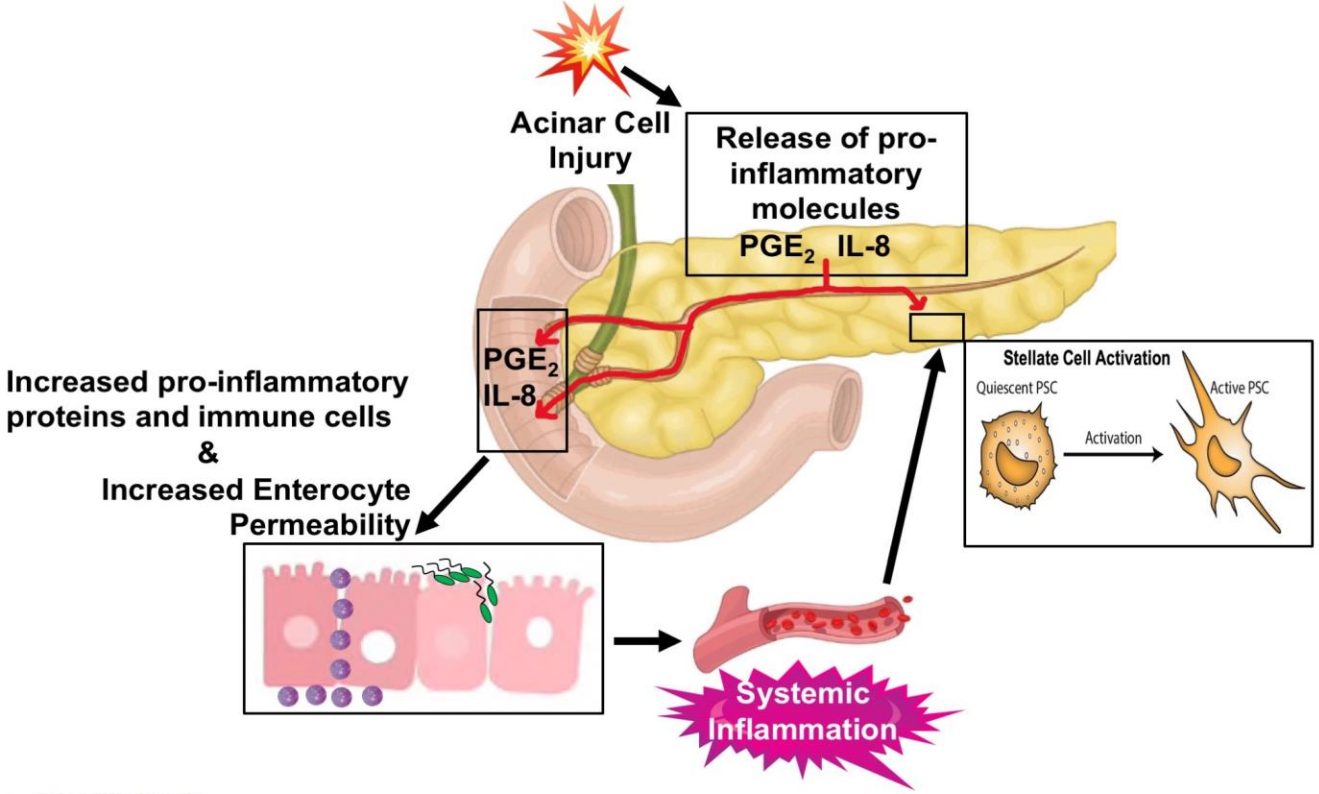


Gastroenterology. 2000; 118: 1051-60

Hypothesis

- PGE_2 within the pancreatic juice of CP patients:
  1. Causes increased intestinal inflammation
  2. Alters duodenal PGE_2 receptor expression, leading to decreased duodenal bicarbonate secretion
  3. Causes impaired intestinal barrier function

Contributes to CP Pathogenesis



Aim 1

To determine the presence of duodenal inflammation and EP receptor and redistribution in CP mice:

- Duodenal tissue (second portion of the duodenum from the Ampulla of Vater to proximal jejunum ~1 cm) fixed and stained by H&E
  - Count Eos, macrophages, neutrophils

- Flow cytometry analysis of isolated duodenal cells from AP, CP, and control mice using eosinophil, macrophage, and neutrophil-specific cell markers (CD3, CD4, CD8, CD16, CD19, CD20, CD160, CD181, CD182)

- qPCR and immunofluorescence measuring EP1-EP4 mRNA and protein distribution in intestine of AP, CP, and control mice

Aim 2

To evaluate duodenal mucosal integrity and bicarbonate secretion in CP mice:

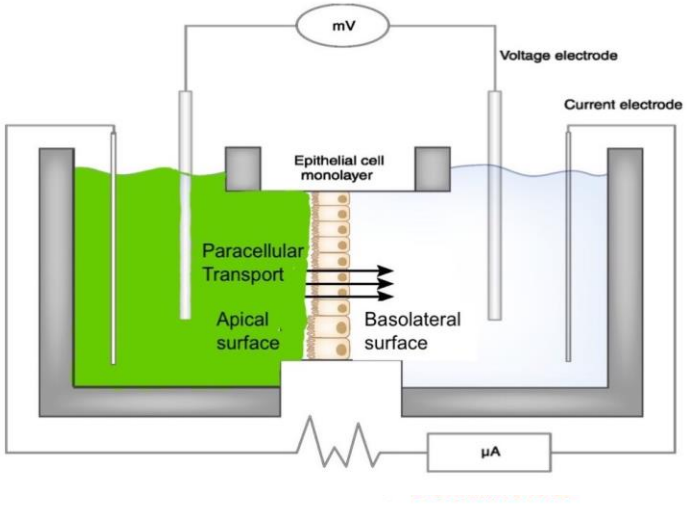
Part A: Impact on Tight Junctions

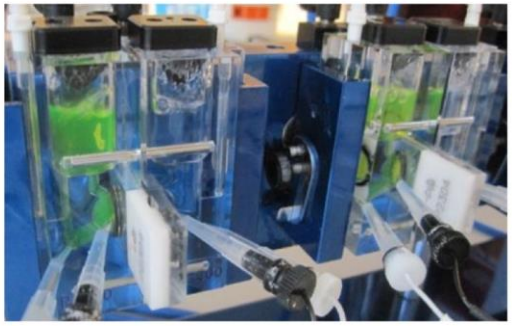
- qPCR target-specific primers to Occludin, Claudins 1-5, ZO-1-3
- Altered expression will be confirmed with Western blot using protein isolates
- Immunohistochemistry will be performed using the same preserved duodenum from Aim 1

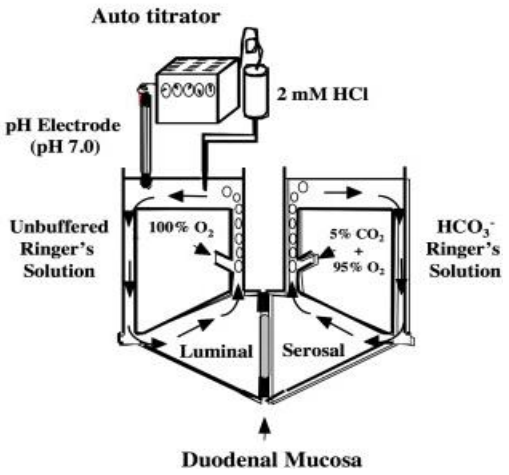
Aim 2

Part B: Investigate functional changes in intestinal barrier function and bicarbonate secretion

- Voltage and current across the duodenal tissue will be measured and transepithelial electrical resistance (TEER) will be calculated using Ohm's Law (V=IxR).
- Simultaneously, bicarbonate secretion is measured by pH-stat
- Fluorescein isothiocyanate-conjugated dextran 4000 (FITC-4000), a large molecule that normally doesn't cross the epithelia, will be added to the mucosal chamber.







Questions?

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