• ERCPs are very helpful, common GI procedures

• Post-ERCP pancreatitis (PEP) is still an important problem

What are the mechanisms underlying the development of post-ERCP pancreatitis (PEP)?
Current understanding about the pathogenesis of PEP

• Exposure of the pancreatic duct to

  1. Mechanical factors—Pressure

  2. Chemical factors—Radiocontrast (RC)
Insights about the pathogenesis of PEP from the current preventatives

• Pancreatic duct stents (relieve pressure)
• Use of a guide-wire (avoid PD injection)
• Rectal indomethacin
  – Why indomethacin?
    • NSAID. Dampens the inflammatory response?
  – Why is the rectal route efficacious?
    • Localized delivery to the region of the pancreas?

• The molecular mechanisms underlying PEP are unclear
Are experimental models of PEP useful for examining the pathogenesis of PEP?

Previously published animal models of PEP

• **Dogs**
  – Comp Med. 2009 Feb;59(1):78-82.

• **Rats**
There an advantage to a mouse model of PEP

- Less expensive

- Genetically engineered mouse models (GEMMS)

- Faster results

- Can be used as an *in vivo* next step after *ex vivo* studies in mouse or human pancreatic cells
PEP model in mice

Gastroenterology. 2015 Sep;149(3):753-64.
Intra-ductal infusion of radiocontrast (RC) at high pressure induces pancreatitis.

Radiocontrast (RC) + high pressure induces an increase in Serum Amylase and Histological Severity compared to Normal Saline (NS).

*From Gastroenterology. 2015 Sep;149(3):753-64.*
What is the role of radiocontrast exposure and pressure on the pancreas in the development of PEP?
Both increased pressure and exposure to radiocontrast contribute to pancreatitis severity.

**Histological Severity**

- In vivo

Both increased pressure and exposure to radiocontrast contribute to pancreatitis severity.

Radiocontrast: - - + +

~Pressure: X 4X X 4X

**Gastroenterology. 2015 Sep;149(3):753-64.**
What is radiocontrast (RC)?

<table>
<thead>
<tr>
<th>Nonionic</th>
<th>Ionic</th>
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<tbody>
<tr>
<td>Monomeric</td>
<td>Monomeric</td>
</tr>
<tr>
<td>iohexol</td>
<td>iopamidol</td>
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</table>
Schematic of the estimated RC concentration exposed to the pancreas during ERCP

10-30% final conc.
Aberrant Ca\textsuperscript{2+} signals within the pancreatic acinar cell are necessary for pathologic events in pancreatitis

Does RC exposure induce Ca\textsuperscript{2+} signals?

If so, are these signals unique to pancreatic cells?
In mouse acini

Radiocontrast (RC) induces acinar cell Ca\(^{2+}\) signals

*Gastroenterology. 2015 Sep;149(3):753-64.*
Ca\textsuperscript{2+} targets in pancreatitis

- Dozens of putative targets
- Which are relevant?

- Calpains
- Calmodulin
- MPTP
- Kinases
- Phosphatase—Calcineurin
Calcineurin (Cn)

**Activators**
- $\text{Ca}^{2+}$
- Calmodulin (CaM)

**Inhibitors**
- FK506, CsA

**Targets**
- NFAT, several others

Does RC exposure activate calcineurin (Cn)?
In mouse acini

RC causes Cn activation

Gastroenterology. 2015 Sep;149(3):753-64.
In AR42J cells

RC causes NF-kB activation, and it is dependent upon Cn

Gastroenterology. 2015 Sep;149(3):753-64.
In mouse acini, RC causes cell injury, that is dependent upon Cn.
What are the effects of Cn on PEP *in vivo*?

*(in which there is the impact of both RC exposure and pressure on the PD)*
In vivo

Cn mediates PEP: Cn inhibitors

NS + low pressure  RC + high pressure  + FK506 (IP)

Histological Severity

Serum Amylase

Gastroenterology. 2015 Sep;149(3):753-64.
CnAβ-deficient mice protected against PEP

Histological Severity

Gastroenterology. 2015 Sep;149(3):753-64.
Is pancreatic acinar cell calcineurin necessary for PEP in vivo?
Acinar cell calcineurin mediates PEP: Ela-Cre^{ERT2}/CnB1^{f/f}

NS + low pressure

CnB1^{flox/flox}  CnB1^{Δ/Δ}

RC + high pressure

CnB1^{flox/flox}  CnB1^{Δ/Δ}

Histological Severity

Score

<table>
<thead>
<tr>
<th></th>
<th>CnB1^{flox/flox}</th>
<th>CnB1^{Δ/Δ}</th>
</tr>
</thead>
<tbody>
<tr>
<td>NS + low pressure</td>
<td>![Bar chart]</td>
<td>![Bar chart]</td>
</tr>
</tbody>
</table>

Orabi & Wen, 2016
Acinar cell calcineurin mediates PEP: AAV6-Ela-Cre in CnB1^{flox/flox} mice

Orabi & Wen, 2016
Clinically target acinar Cn in vivo during PEP?

Would a single acute dose of Cn inhibitors within the radiocontrast infusion mitigate PEP?
Intraductal (ID) administration of calcineurin inhibitors prevents PEP.

Orabi & Wen, 2016
Intraductal FK506 prevents both pancreatic and serum IL-6 elevations during PEP

Pancreatic IL-6

Serum IL-6
Healthy skepticism

• All models have limitations

• How relevant is the PEP model in mice to man?
• Will the RC/Cn formulation be useful in man?

• Large animal confirmation, and then first in man studies
Summary

• Experimental models of PEP are useful for deciphering the mechanisms underlying PEP

• A novel model of PEP in mice offers several advantages

• PEP can be mimicked by a combination of RC exposure and elevated pressure on the pancreas

• $\text{Ca}^{2+}/\text{Cn}$ is a critical signaling pathway in PEP

• A novel RC/Cn formulation could be useful in preventing PEP
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More questions about RC and the novel formulation

• Will the novel RC/Cn formulation be useful in ERCP cases where RC is not injected into the pancreatic duct?

• What is the utility of the RC formulation on pressure-related PEP without RC exposure?

• What is the effect of RC (and similarly Cn blockade due to the novel RC formulation) on the other components of the pancreas?
  – Whole pancreas, duct cells?
  – PD studies. Also, PK studies.

• How does the RC trigger Ca$^{2+}$ in the acinar cell and initiate inflammatory change?
  – Building a better RC?

• Are there calcineurin-independent or at least contributing pathways in PEP that could be targeted within the RC formulation?
  – Building a better RC formulation?

• Hence the need for experimental models of PEP