Pancreatic Cancer: Bench to Bedside

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Pancreatic Cancer in 2016

- ~45,000 new cases diagnosed each year in the U.S.
  - 4th leading cause of cancer-related death
  - 85% ductal adenocarcinoma
  - Nearly all patients will die of their disease

- Surgical resection is the only potential cure

- Only 15-20% patients are candidates for resection
  - 5-year survival 25-30% for node-negative tumors
  - 5-year survival 10% for node-positive tumors
Pancreatic Cancer is Projected to Become the 2nd Leading Cause of Cancer Death by 2020

Projected cancer deaths (thousands)

2010 2020 2030

lung pancreas colorectal breast prostate

Reproduced from the Pancreatic Cancer Action Network.

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Pancreatic Cancer Survival of 3 Decades

5-year Relative Survival Rate

<table>
<thead>
<tr>
<th>Year of diagnosis</th>
<th>Percentage diagnosed with invasive pancreatic cancer</th>
</tr>
</thead>
<tbody>
<tr>
<td>1975-1977</td>
<td>2.4%</td>
</tr>
<tr>
<td>1984-1986</td>
<td>2.9%</td>
</tr>
<tr>
<td>1996-1998</td>
<td>4.4%</td>
</tr>
<tr>
<td>2002-2008</td>
<td>6.0%</td>
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</tbody>
</table>

Pancreatic Cancer is Detected in Late Stages and Associated with a Poor Prognosis

Stage at Diagnosis 2003-2009

- Localized: 53%
- Regional: 27%
- Metastatic: 9%
- Unknown: 11%

5-Year Relative Survival 2003-2009

- Localized: 24.1%
- Regional: 9.0%
- Metastatic: 2.0%
- Unknown: 4.1%

Personalized Medicine Approach to Pancreatic Cancer

- Currently only one available personalized medicine approach: DNA sequencing
  - Limited availability
  - Limited benefit for most patients

- Pancreatic cancer xenografts cannot be generated quickly enough to benefit patients

- Rapid generation of novel, personalized cancer models are desperately needed
Pancreatic Cancer ORGANOIDS

• “Organoids”
  – 3D organ-bud grown \textit{in vitro} from healthy tissue
  – Simulate the micro-environment of the organ
  – First reported with healthy, intestinal crypt cells
    • Sato T and Clevers H, \textit{Nature} 2009

• Pancreatic Cancer Organoids
  – Generated from human tumors
  – Reproduce the full spectrum of tumor development
  – Exhibit ductal and disease-stage specific characteristics
  – First reported in 2015
    • Boj SF and Tuveson D, \textit{Cell} 2015
Human Normal and Neoplastic ORGANOIDs
Generated from Surgical Resection Specimens

PDA Surgical Tumor Resection

Small normal specimen selected by Pathology
Sample arrives ~4 - 5 hr after surgery
2 - 4 hr digestion Collagenase TrypLE
Plate normal cell mixture in Matrigel

Small tumor specimen selected by Pathology
Sample arrives ~4 - 5 hr after surgery
4 hr - overnight digestion Collagenase TrypLE
Plate tumor cell mixture in Matrigel

Boj et al, Cell 2015
Human Normal and Neoplastic ORGANOIDs
Generated from Surgical Resection Specimens

PDA Surgical Tumor Resection

Small normal specimen selected by Pathology
Sample arrives ~4 - 5 hr after surgery
2 - 4 hr digestion Collagenase TrypLE

Tumor Resection

Small tumor specimen selected by Pathology
Sample arrives ~4 - 5 hr after surgery
4 hr - overnight digestion Collagenase TrypLE

Plate normal cell mixture in Matrigel
Plate tumor cell mixture in Matrigel

Primary or Metastasis Fine Needle Biopsy

FNA biopsy arrives ~4 - 5 hr after the procedure
Red blood cell lysis
No digestion

Plate cell mixture in Matrigel

Boj et al, Cell 2015
EUS-guided Fine-Needle Biopsy (EUS-FNB) for Tissue Diagnosis & Organoid Creation

video
ORGANOID Isolates from Limited FNB Material

Primary or Metastasis
Fine Needle Biopsy

FNA biopsy arrives
~4 - 5 hr after
the procedure

Red blood cell
lysis

No digestion

Plate cell
mixture in Matrigel

hF2

hF2
Human Neoplastic **ORGANOIDS**
Generated from EUS-FNB Specimens

<table>
<thead>
<tr>
<th>Organoid</th>
<th>Brightfield</th>
<th>H&amp;E</th>
<th>Tissue</th>
<th>H&amp;E</th>
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</thead>
<tbody>
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<td>hN1</td>
<td><img src="image1.png" alt="Image" /></td>
<td><img src="image2.png" alt="Image" /></td>
<td><img src="image3.png" alt="Image" /></td>
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<tr>
<td>hFNA2</td>
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<td><img src="image22.png" alt="Image" /></td>
<td><img src="image23.png" alt="Image" /></td>
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Collaborative Research Effort

Stony Brook Medicine

Cold Spring Harbor Laboratory

SUNY Downstate Medical Center

American Society for Gastrointestinal Endoscopy

2015 Endoscopic Research Award
EUS-FNB for Creation of Pancreatic ORGANOIDS At the Time of Initial Diagnosis

• Current status:
  – 26 patients enrolled
  – Only 22% went for subsequent surgical resection
  – **Successful organoid creation within 2 weeks of FNB in 22/26 (85%)**
  – 2 patients who underwent surgical resection had successful matching between FNB organoid & surgical resection organoid
  – 3 patients initiated *in vitro* drug sensitivity testing using their FNB organoid specimen
Effect of gemcitabine on human pancreas organoids

% viability

Log drug concentration

hF2
hM1A
hM6
hT2
hT3
hM1E
hF23
hF24

Gemcitabine Testing
Paclitaxel Testing

Effect of paclitaxel on human pancreas organoids

% viability

Log drug concentration
Conclusions

• Pancreatic cancer survival rates remain exceptionally low
• Most patients are not candidates for curative resection
• EUS specialists are uniquely positioned to interact with patients at the time of initial tissue diagnosis
  – Optimal time for cultivating methods that allow for future personalized treatment options
• Pancreatic cancer organoids can be generated rapidly from EUS-FNB specimens
  – Especially important for the vast majority not going for surgical resection
• Organoids serve as the framework for precision medicine
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