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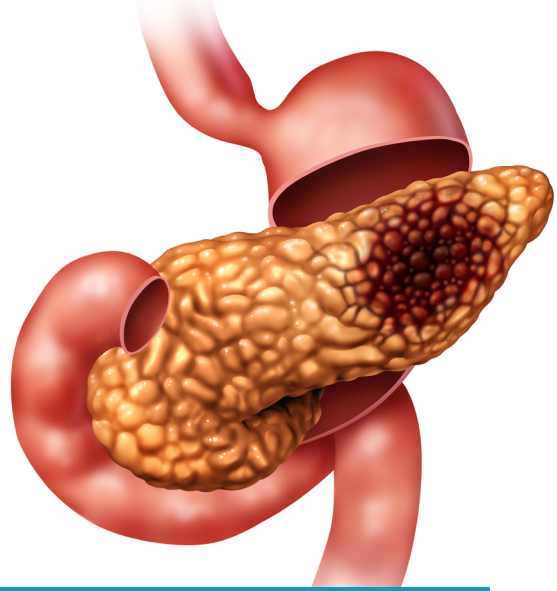


In Vivo Microscopy for the Evaluation of Pancreatic Lesions

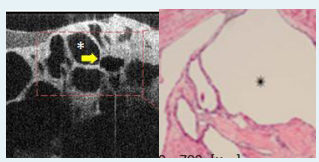
In vivo microscopy (IVM) applies light directly to living tissues to produce images that may be interpreted by a trained pathologist for diagnosis. One important application is imaging of pancreatic lesions via endoscopic procedures.

Overview of In-vivo Microscopy Technologies Used for its Evaluation

Optical Coherence Topography (OCT)

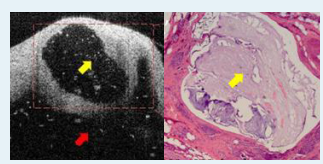
- **OCT-based IVM provides ultra-sound like cross sectional images at microscopy resolutions (up to a few microns)**
- **OCT provides information about cyst architecture and content for diagnosis of serous and mucinous cysts**

OCT Images



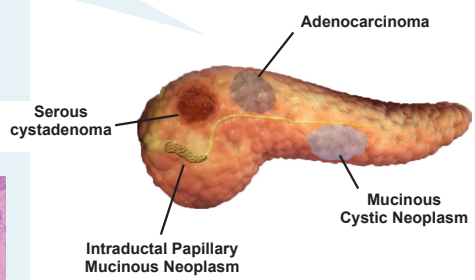
Honeycomb appearance. Dark cystic spaces divided by white thick septae[1]

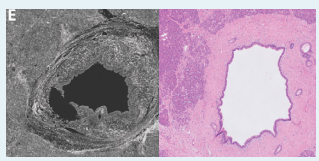
Serous cystadenoma[1]



Well-circumscribed dark cystic space with some scattering[1]

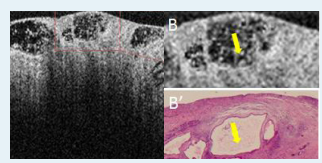
Mucinous cystic neoplasm[1]





Normal pancreatic duct and surrounding parenchyma[3]

Normal pancreatic duct with surrounding parenchyma[3]



Multiple cysts with highly scattering mucin[1]

IPMN[1]

Assessment of Cystic Pancreatic Strictures

- Ex-vivo studies show the ability of OCT in differentiating mucinous from serous content of cysts based on the scattering properties of the cyst contents
- Cross sectional imaging using OCT allows for assessment of cyst size, wall thickness and extent of septation for differentiation of serous and mucinous cysts

Assessment of Malignant Pancreaticobiliary Strictures

- OCT probes may be passed through the main pancreatic duct and bile duct during endoscopic procedures for the diagnosis of malignant strictures
- OCT of a normal main pancreatic duct shows a tri-layer architecture representing the epithelial lining, fibromuscular layer surrounding the epithelium and loose connective tissue / acini surround them[3]
- Adenocarcinoma involving the main pancreatic duct shows loss of the tri-layered architecture and multiple minute non-reflective areas seen on OCT[3]
- OCT can detect main pancreatic duct adenocarcinoma with an accuracy approaching 100%[3]

Needle-Based Confocal Endomicroscopy (nCLE)

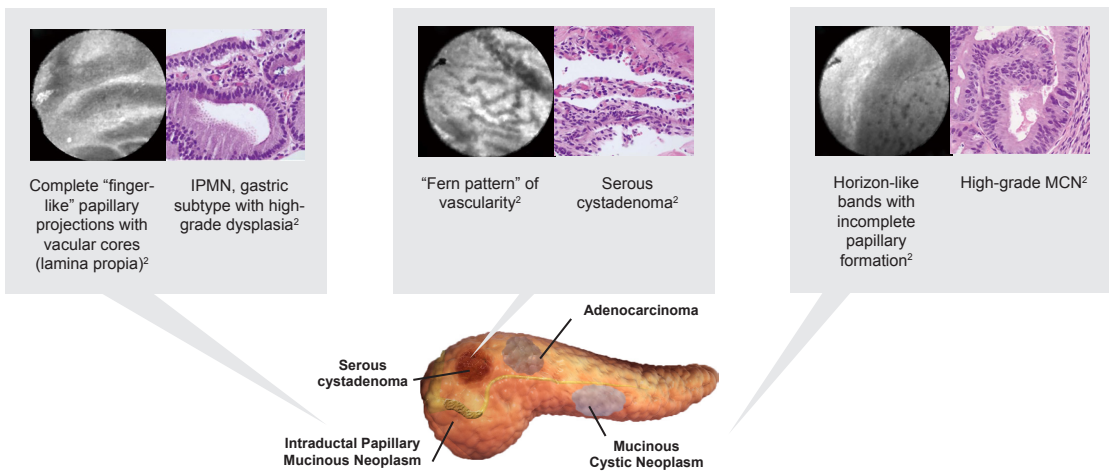
- **In needle-based endomicroscopy, an optical probe is passed through a 19-gauge needle during endoscopic ultrasound, fine needle aspiration (EUS-FNA) procedures and produces images in concert with injected Fluorescein contrast.**
- **The probe is placed directly against the cyst wall.**
- **Vascular and epithelial fluorescence and patterns allow for diagnosis of various pancreatic lesions.**

Fluorescent Agents Used in CLE

Appearance of tissue using CLE often depends on the contrast agent used.

- The widespread use of injectable contrast agents such as fluorescein has led to the development of diagnostic criteria based on vascular architecture and hemo-dynamics.
- "Leaky vessels" showing leakage of fluorescent agents usually signify malignancy.
- Topical fluorescent agents such as acriflavine and proflavine stain nuclei allow for assessment of nuclear morphology, distribution and size analogous to classic histopathology.
- Research in in-vivo molecular fluoroprobes that are specific for pancreatic adenocarcinoma may increase sensitivity and specificity of in-vivo diagnoses using CLE.[1]

CLE Images



Assessment of Cystic Pancreatic Structures

- Papillary structures are easily identified using probe-based CLE in intraductal papillary mucinous Neoplas
- CLE probes allow the visualization of a superficial vascular network in the cyst walls of serous cystadenomas, a sign that is highly specific for the disease

Assessment of Malignant Pancreaticobiliary Structures

- CLE probes may be passed through the main pancreatic duct and bile duct during endoscopic procedures for the diagnosis of malignant strictures
- Criteria suggestive of a malignant stricture involving a pancreatic or biliary duct include:
  - o Thick dark bands (>40 microns)[2, 3, 4]
  - o Thick white bands (>20 microns) [2, 3, 4]
  - o Dark Clumps[2, 3, 4]
  - o Fluorescein leakage2, 3, 4
- CLE predicts neoplasia involving the pancreaticobiliary ducts with a sensitivity and specificity of 83% and 75% (compared to 65% and 53% using cytology), respectively[3]

Text References

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Image References

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3. van Manen L. Stegehuis PL, Farina-Sarasqueta A, de Haan LM, Eggermont j, Bonsing BA, et al. (2017) Validation of full-field optical coherence tomography in distinguishing malignant and benign tissue in resected pancreatic cancer specimens. PLoS ONE 12(4): e0175862. For information about the open access license, please refer to http://journals.plos.org/plosone/s/licenses-and-copyright.

*For the Iftimia images without captions: Biomed. Opt. Express 2, 2372 (2011)