Pancreatic Cytopathology: A pragmatic approach.

By
Dr Miguel Perez-Machado
MD. PhD. MRCPath
Royal Free Hospital

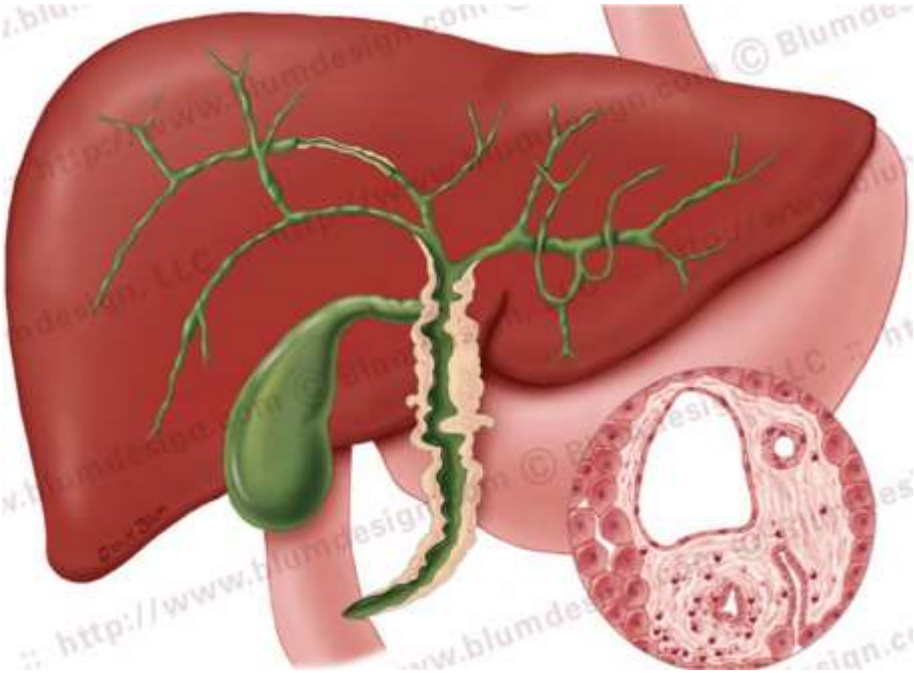
Indications of Cytology Sampling

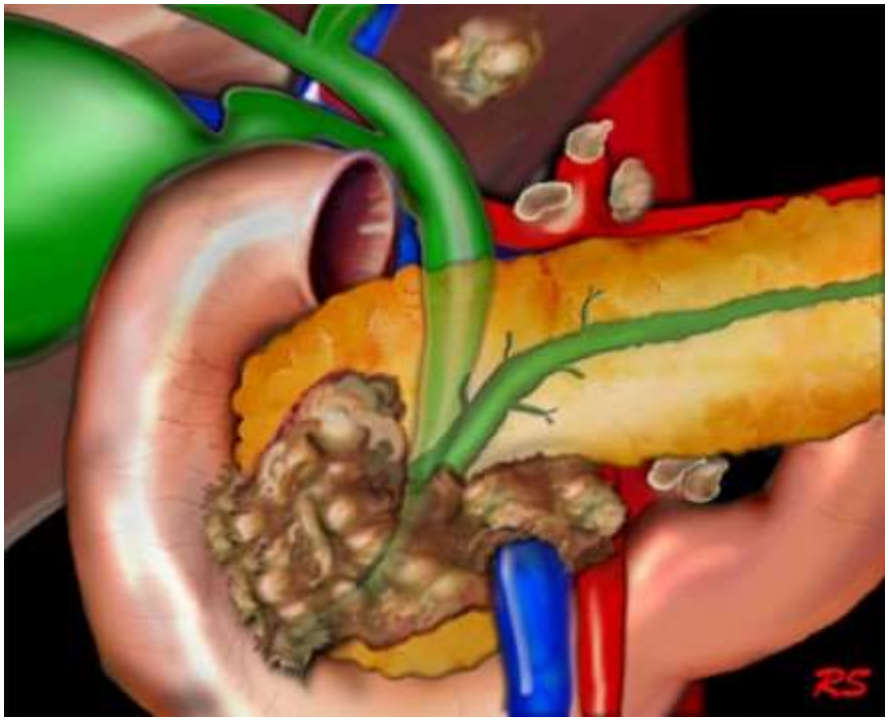
- To document malignancy in patients with malignant – appearing pancreatic masses on imaging.
- If tumour is inoperable, the diagnosis can preclude unnecessary surgery and allow the initiation of chemotherapy and/or radiation therapy
- If tumour is operable, the diagnosis allows for the optimal planning of surgery

Methods of Sampling

For ductal abnormalities (strictures or dilatations) and for interventional capacity (stenting).

For pancreatic masses





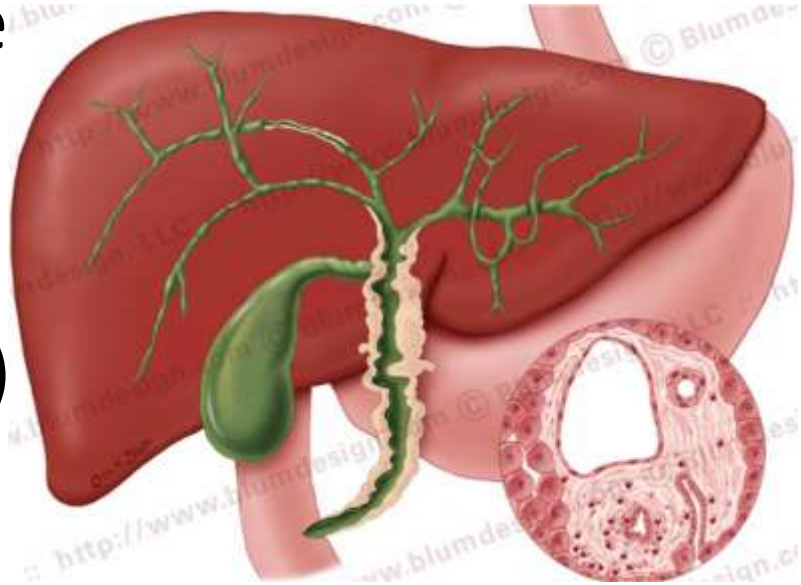
Methods of Sampling

For ductal abnormalities (strictures or dilatations) and for interventional Capacity (stenting).
Do not generally allow for the appreciation of a mass.

- Brushings or aspiration from the within the common bile or pancreatic duct

- ERCP (Endoscopic retrograde cholangiopancreatography)

- PTC (percutaneous transhepatic cholangiography)

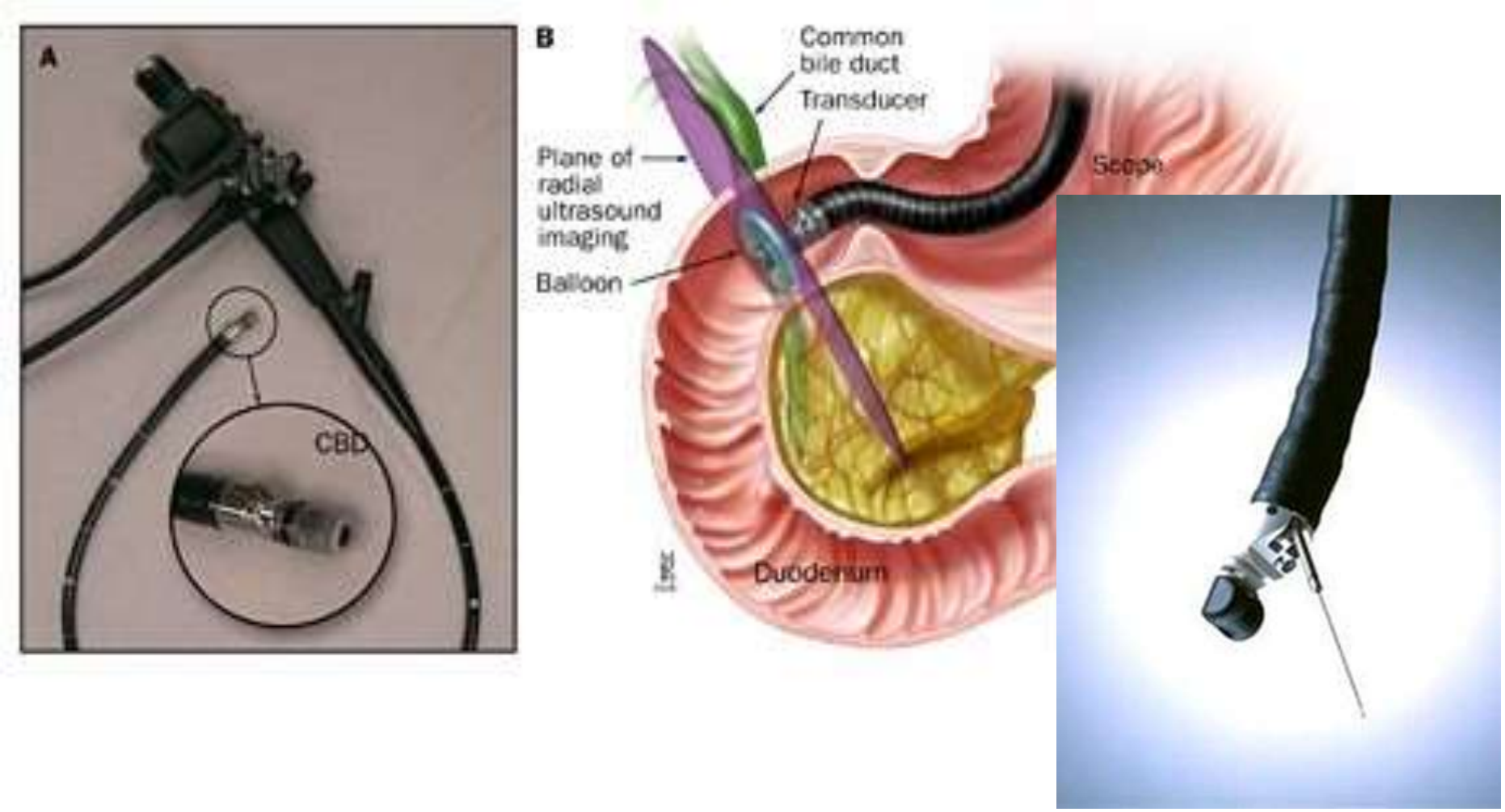


Method of sampling

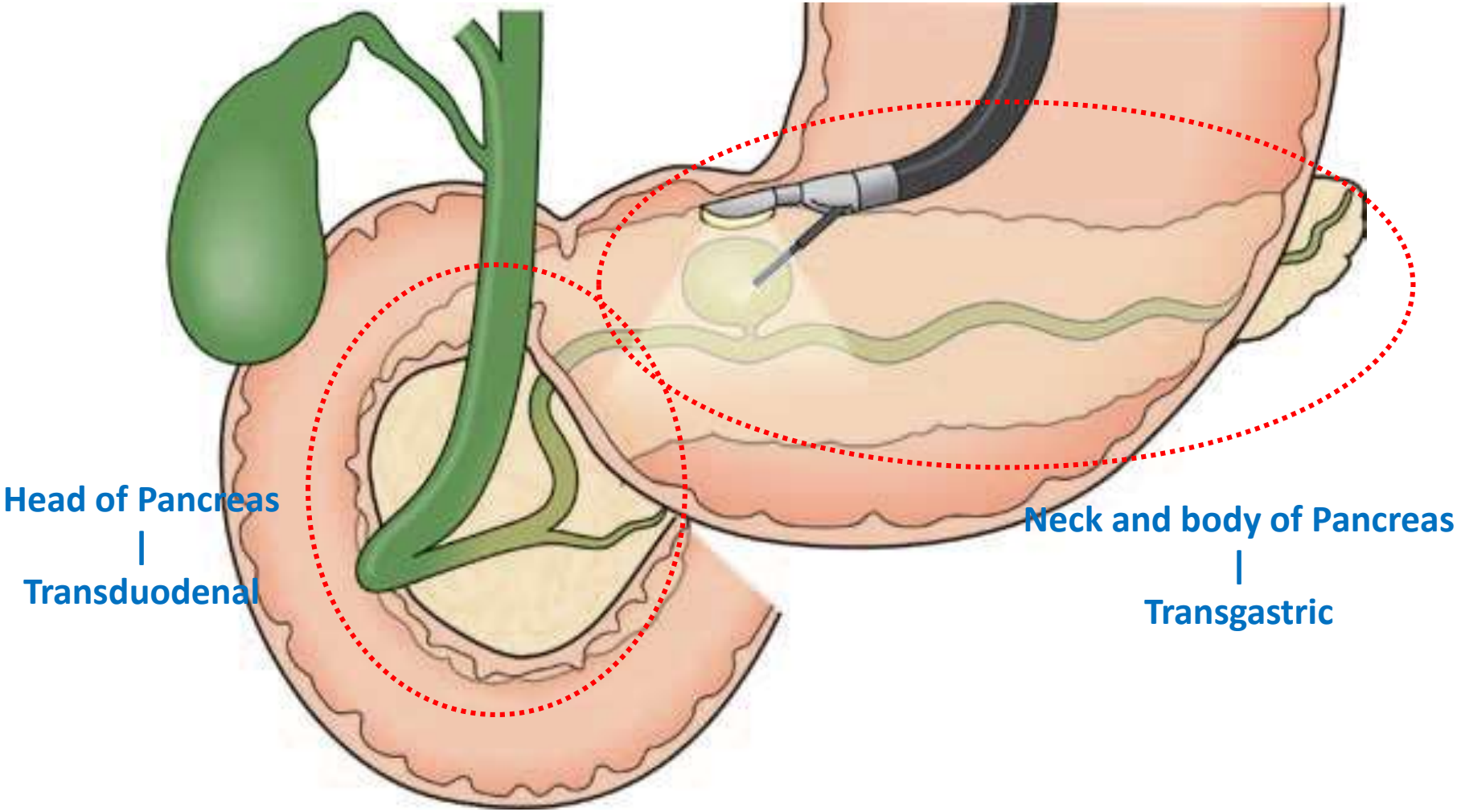
of pancreatic masses

- Aspirate intraoperatively either by palpation or direct visualization.
- EUS-FNA (FNA under endoscopic ultrasound guidance): Allows continuous, real time visualization of a needle as it punctures its target lesion. Ability to sample small lesions, can better detect vascular and nodal involvement.
- Under image guidance
  - Transabdominal ultrasound (US): Allows sampling of the lesion in real time but visualisation can be impaired by bowel and fat.
  - Computed tomography (CT): Better visualisation and resolution, but does not allow for the real time identification of the needle tip during sampling

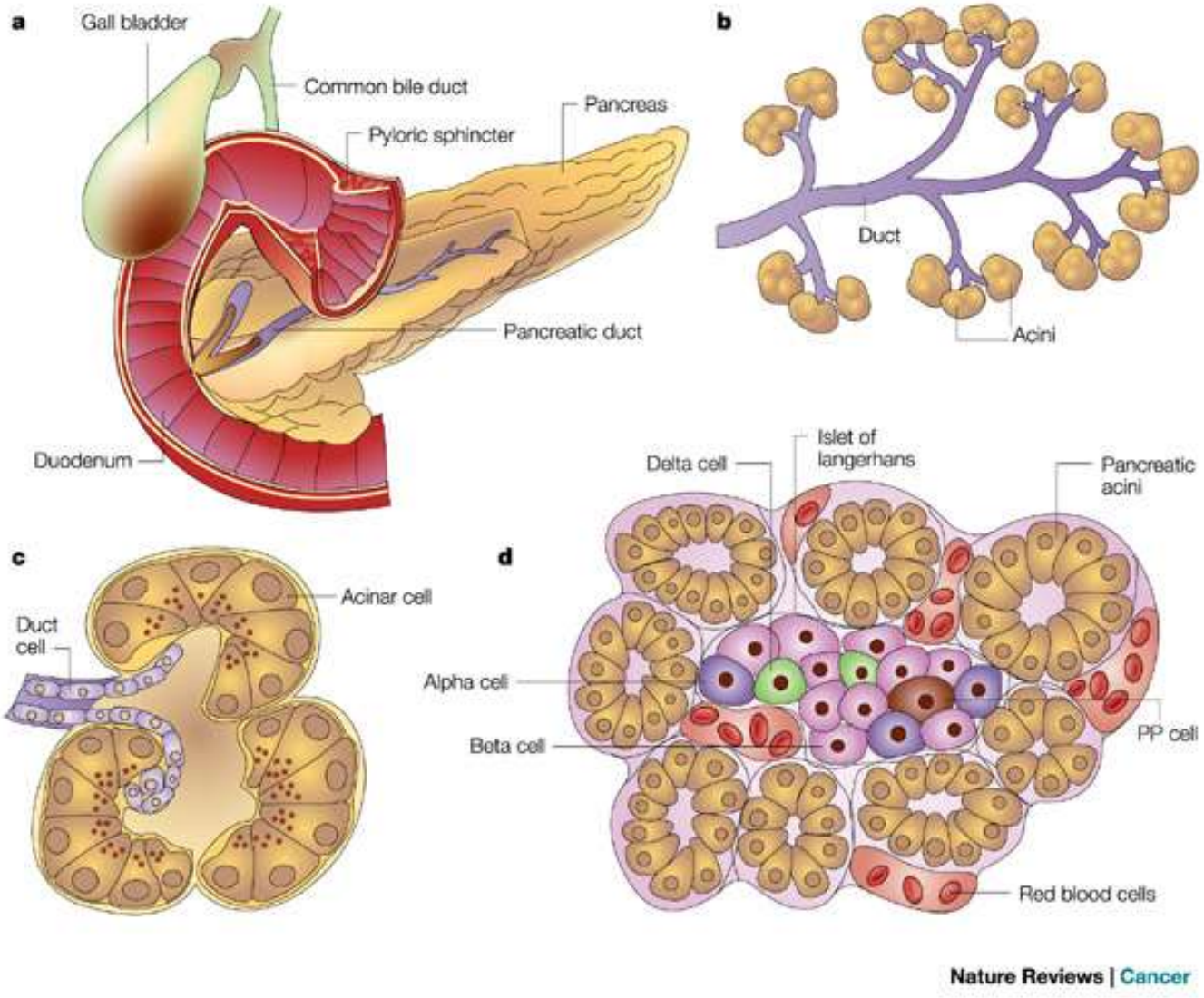
EUS-FNA (FNA under endoscopic ultrasound guidance)



ERCP (Endoscopic retrograde cholangiopancreatography)



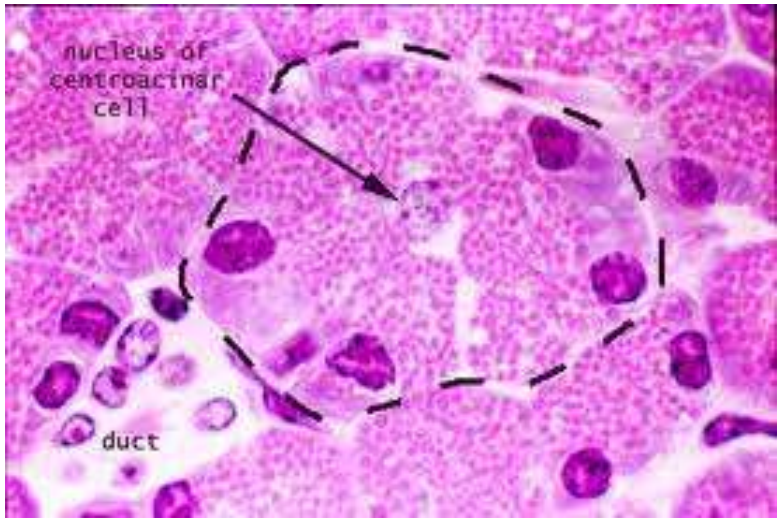
Pancreas anatomy and histology

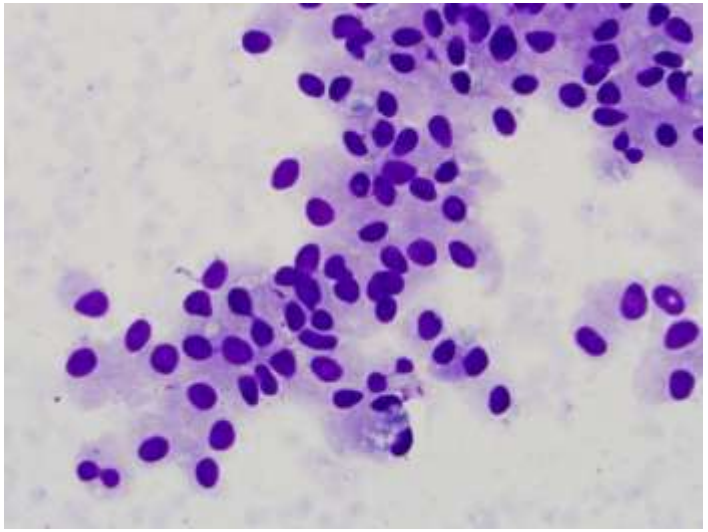


Cytology of normal pancreas

- Acinar cells
  - Predominant cell type
  - Pyramidal or triangular
  - Abundant granular cytoplasm
  - Round, eccentric or central nuclei
  - Fine chromatin and often distinct nucleoli

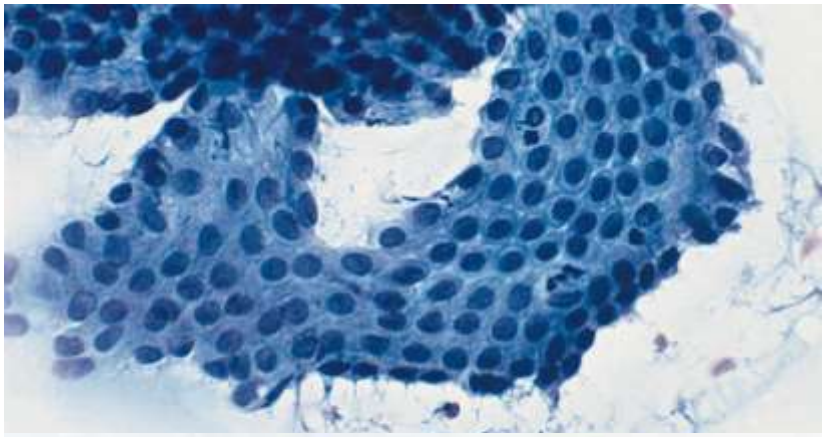
Trypsin +ve

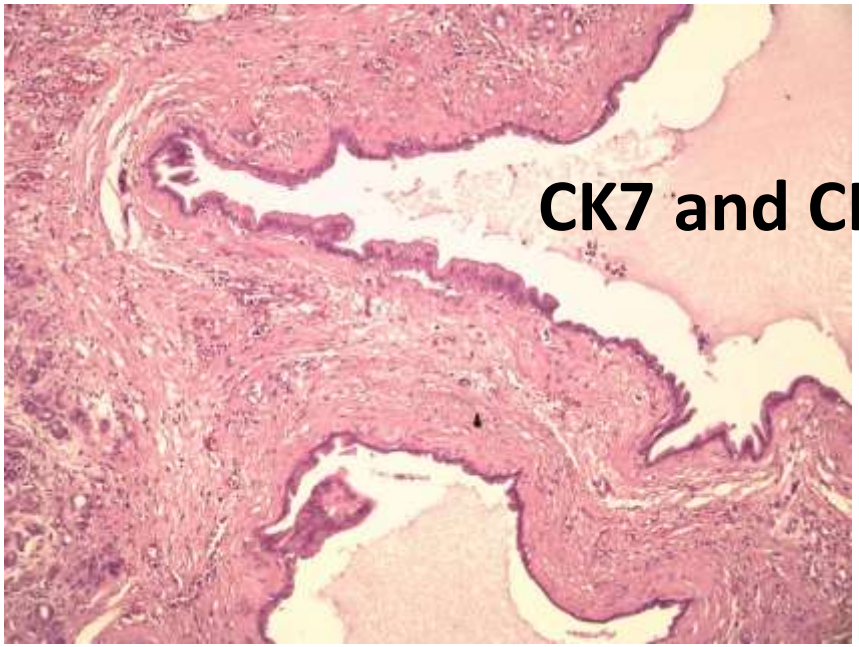




Ductal cells

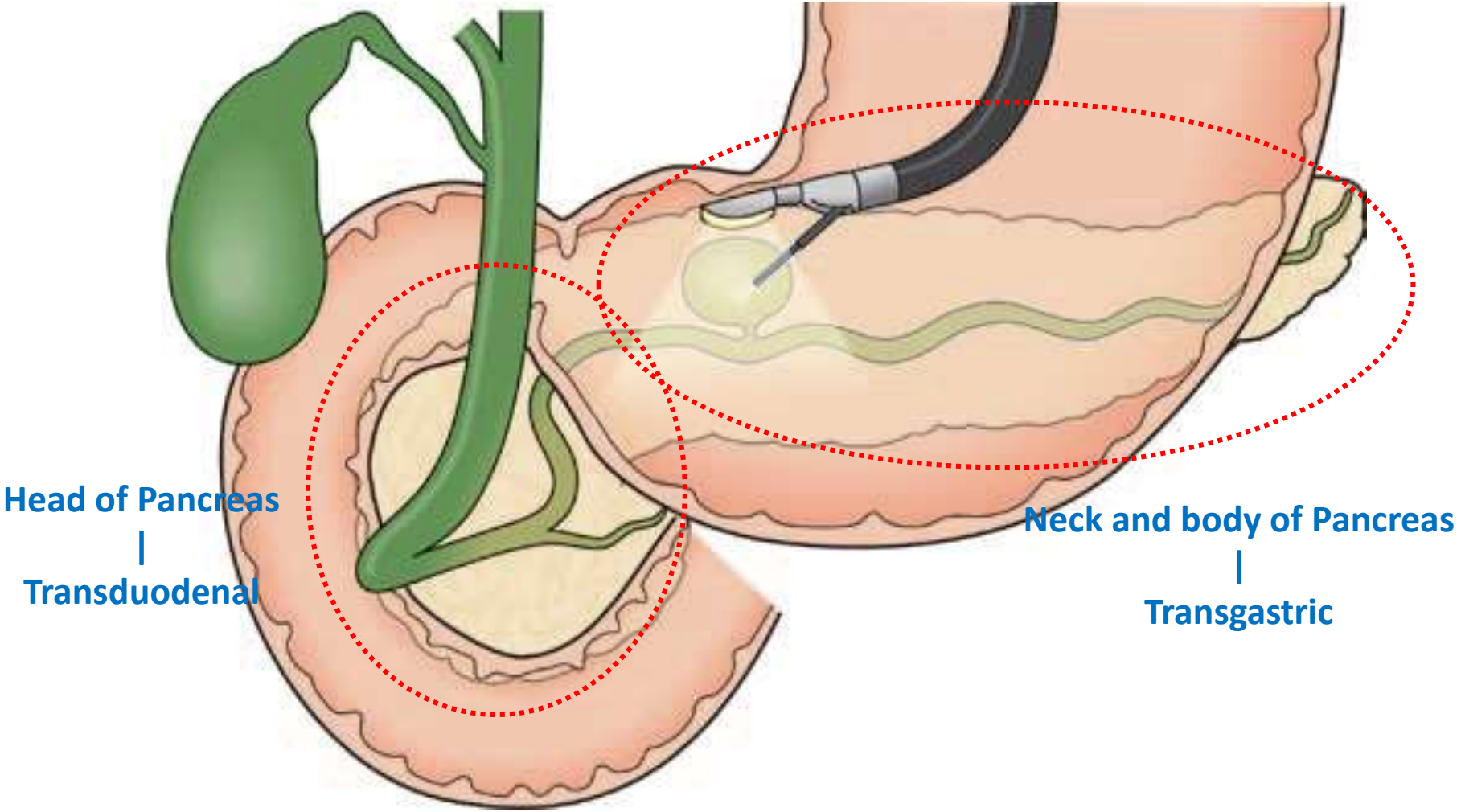
- Two – dimensional flat sheets with “honeycomb” appearance
- “Picked – fence” arrangement with basally located nuclei
- Cuboidal or columnar shaped
- Scant, pale cytoplasm
- Bland appearing nuclei



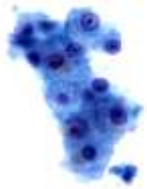
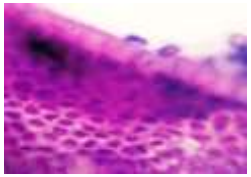


CK7 and CK19 +ve

ERCP (Endoscopic retrograde cholangiopancreatography)



Contaminants

Cell type	Approach	Cytological features
Mesothelial 	Percutaneous	2 – dimensional flat sheets Round to oval nuclei Moderate pale cytoplasm Intercellular windows
Hepatocytes 	Percutaneous	Polygonal cells Abundant, well defined granular cytoplasm Round to oval nuclei Prominent nucleoli +/- cytoplasmic pigment
Bowel mucosa 	Endoscopic Transduodenal (lesion in the pancreatic head and uncinate)	2 – dimensional flat sheets “honeycomb” Round, evenly spaced and bland appearing nuclei Pale cytoplasm with well defined borders Intermixed goblet cells Admixed with extracellular mucus
Gastric mucosa 	Endoscopic Transgastric (lesions in the pancreatic body and tail)	2 – dimensional flat sheets “honeycomb” Round, evenly spaced and bland appearing nuclei Pale cytoplasm with well defined borders Goblet cells are RARE Admixed with mucin

Bowel Duodenal mucosa

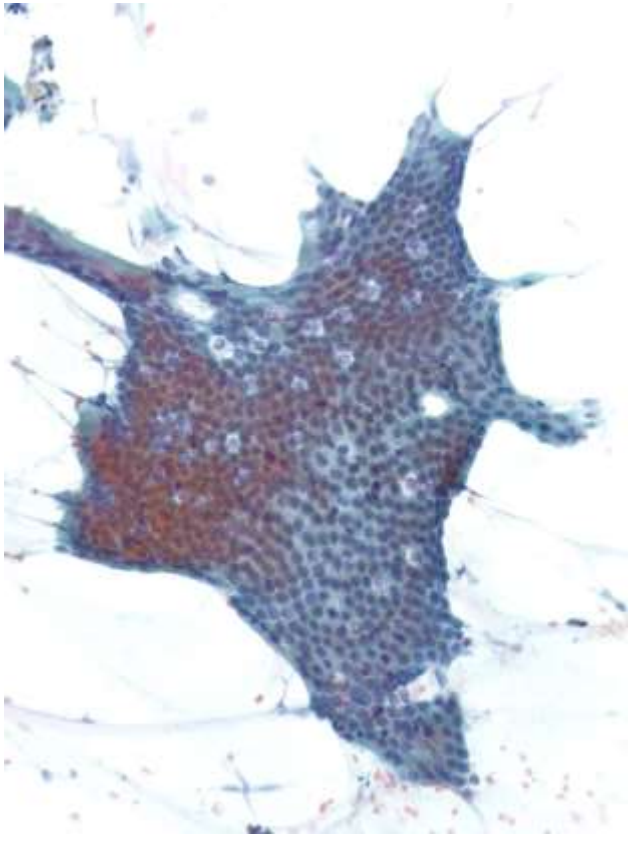
2 – dimensional flat sheets “honeycomb”

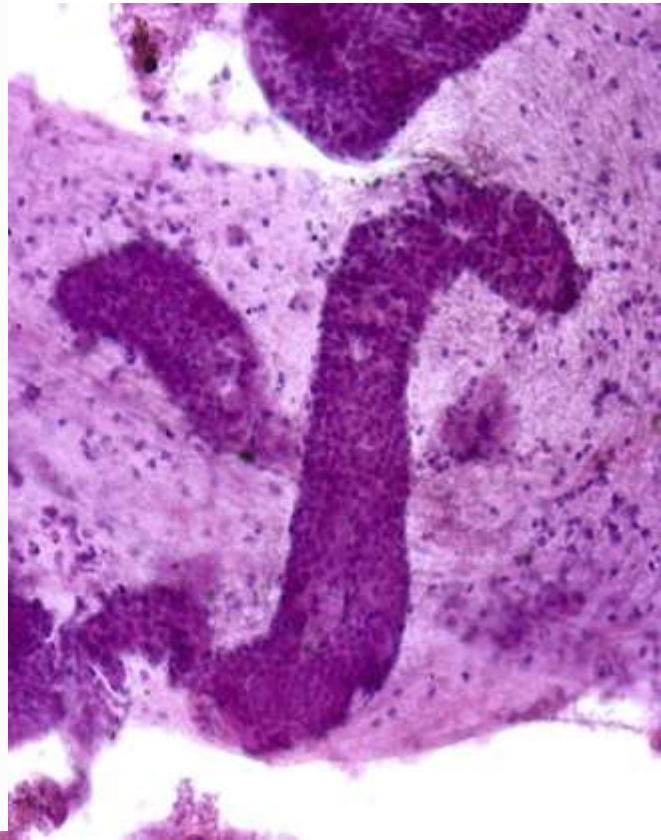
Round, evenly spaced and bland appearing nuclei

Pale cytoplasm with well defined borders

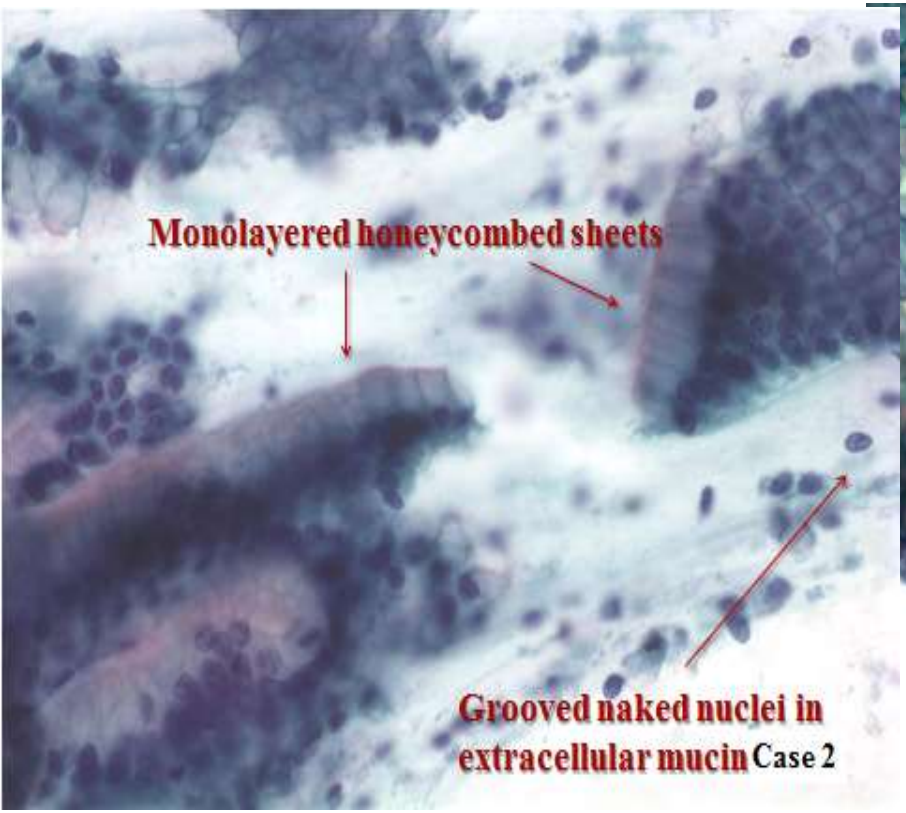
Intermixed goblet cells

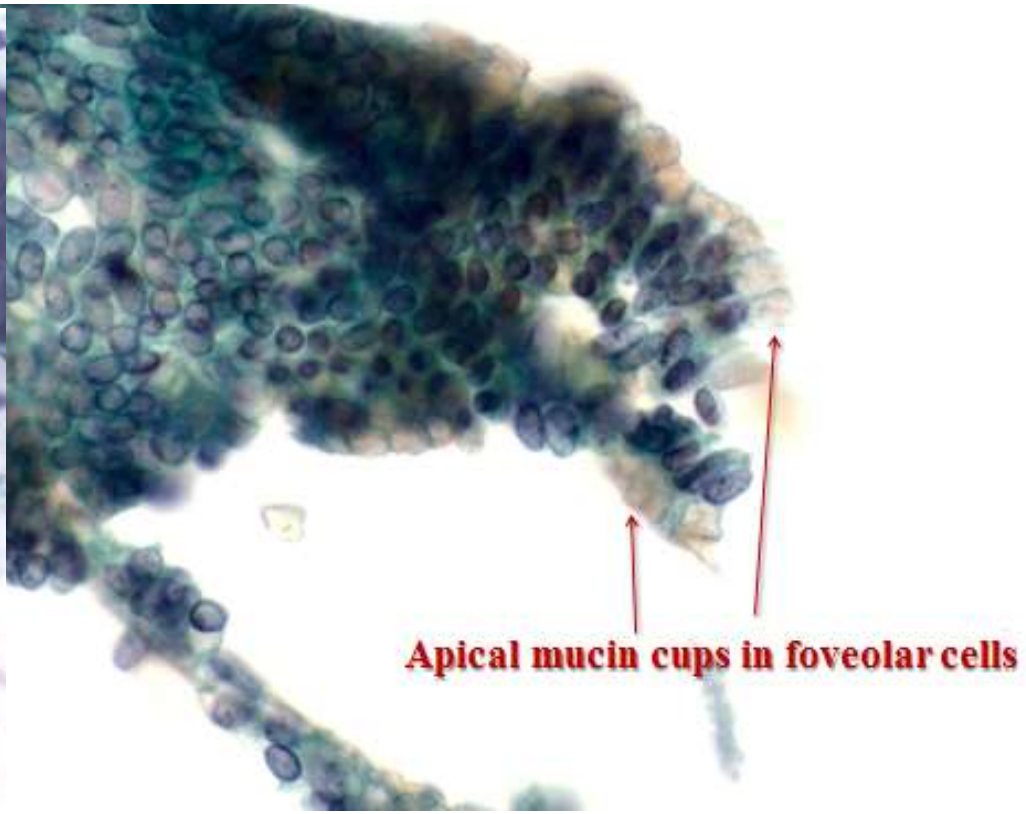
Admixed with extracellular mucus





Gastric mucosa
2 – dimensional flat sheets “honeycomb”
Round, evenly spaced and bland appearing nuclei
Pale cytoplasm with well defined borders
Goblet cells are RARE
Admixed with mucin





Questions to answer before you report your pancreatic cytology

It is solid of cystic?

It is a man or a women?

Age?

Location

a) **Head?**
b) **Neck?**
c) **Body?**

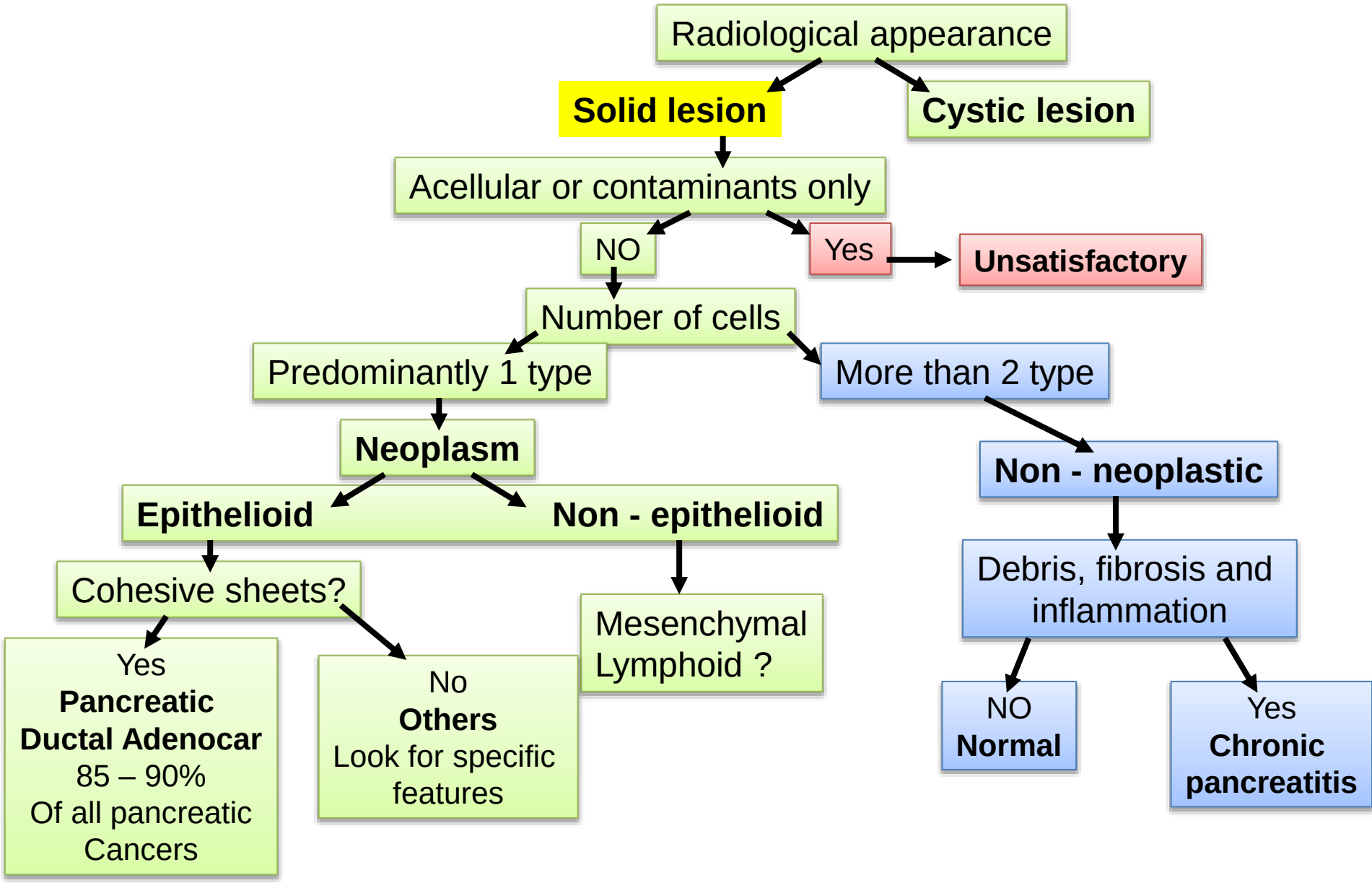
What sort of sampling?

a) **Brushings?**
b) **FNA?**

Any relevant history?

a) **Alcohol abuse?**
b) **PSC?**
c) **Autoimmune pancreatitis?**
d) **Stents?**
e) **Stones?**

Pancreatic Cytopathology. A Pragmatic Approach



The cytology specimen must be assessed taking in consideration the context of patient presentation

Solid mass

Benign / Non-Neoplastic

Pancreatitis (Acute, Chronic, *Autoimmune pancreatitis)*

Ectopic spleen

Neoplastic

Ductal Adenocarcinoma

Neuroendocrine tumours

Acinal cell carcinoma

Pancreatoblastoma

Lymphoma / plasmacytoma

Metastasis

Inflammatory Disease of the Pancreas

Acute Pancreatitis:

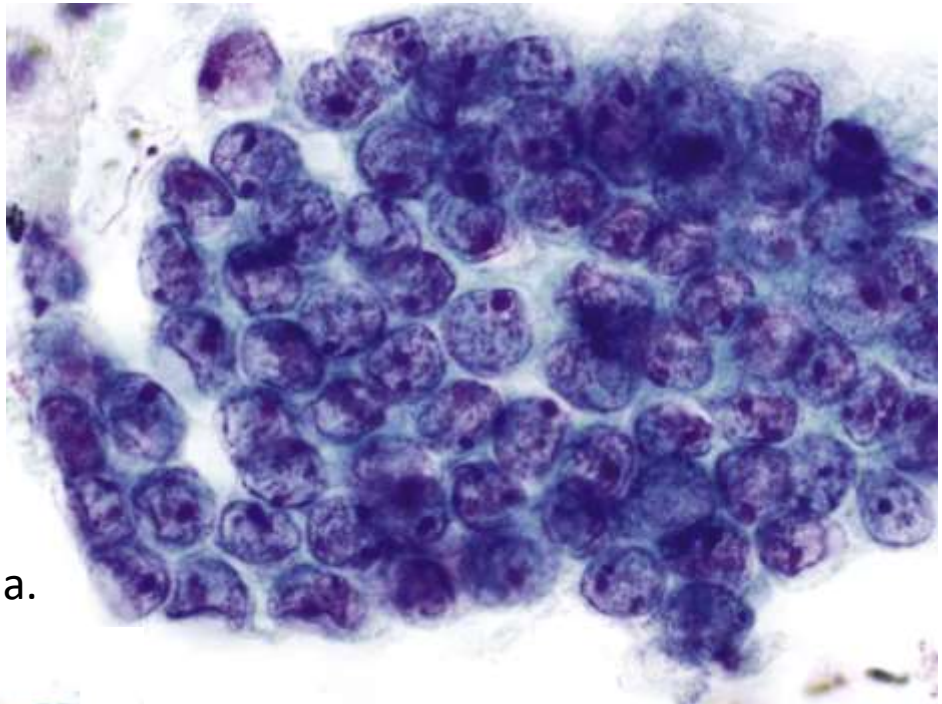
- Characteristic clinical signs and signs and symptoms as well as laboratory findings (↑ amylase and lipase) → No need for tissue diagnosis.

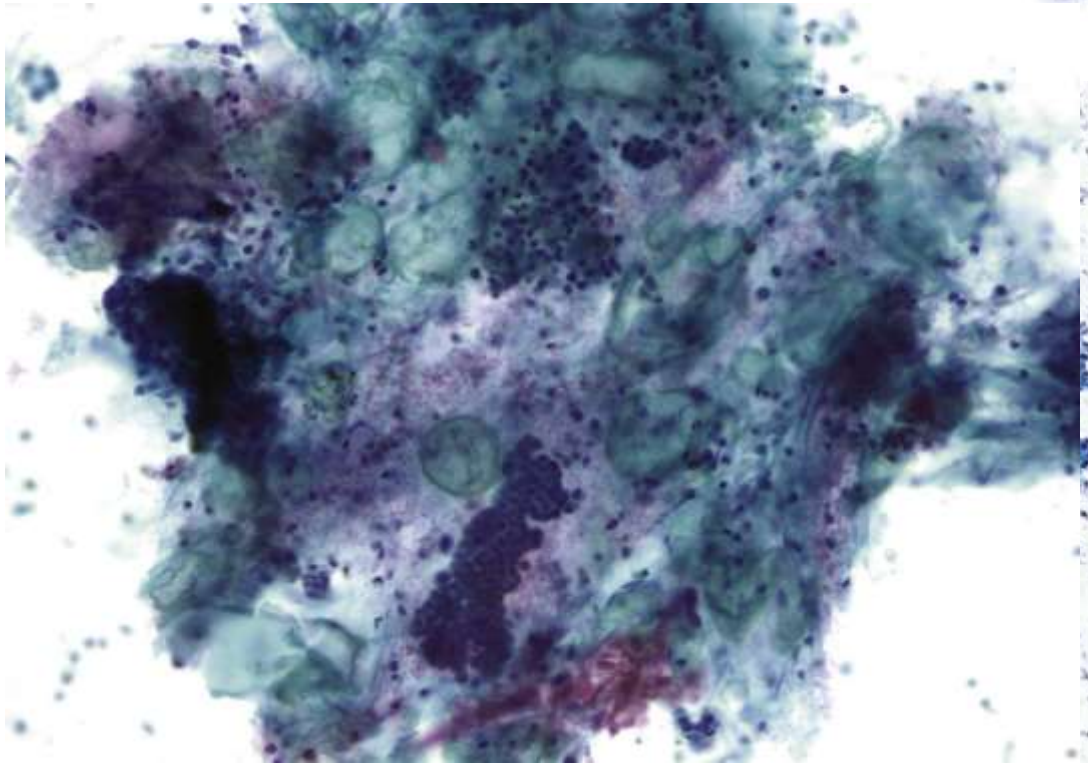
Chronic Pancreatitis:

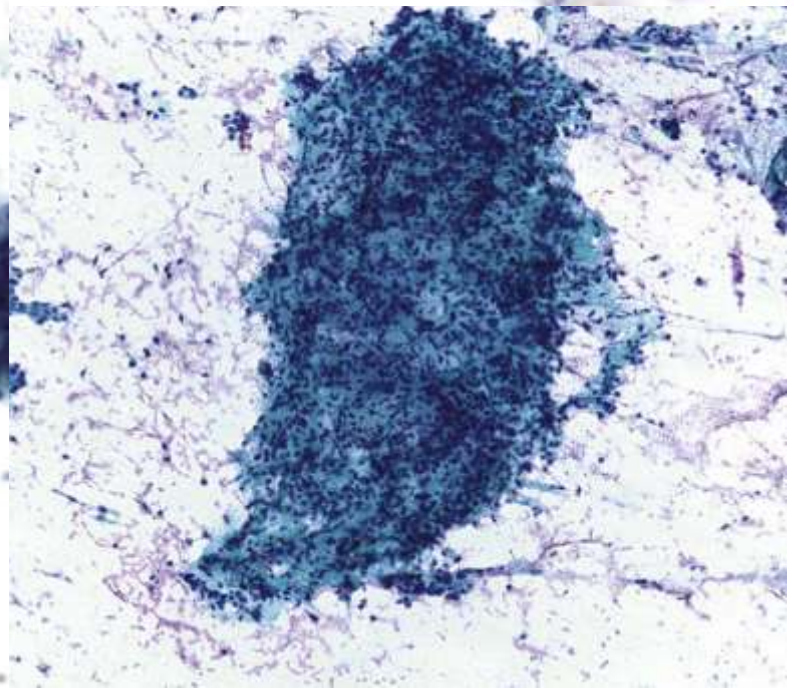
- Insidious
- Clinical and radiological overlap with malignancy and often coexist with Malignancy → Require tissue diagnosis

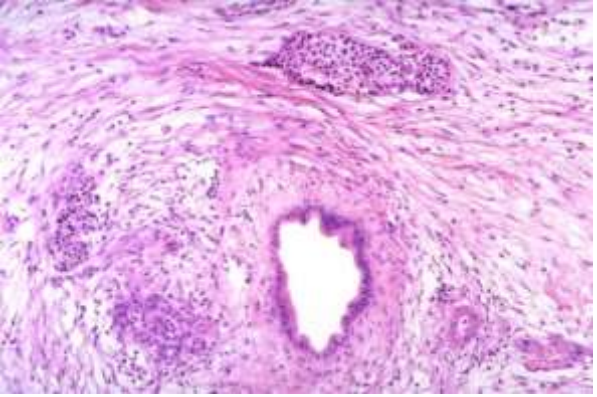
Cytological Features of Chronic Pancreatitis

Background composed of grungy material and calcification
Fibrotic stroma tissue
Fibrotically distended acinar tissue
Inflammation, mixed, usually not severe
Pancreatic elements with mild cytological atypia.



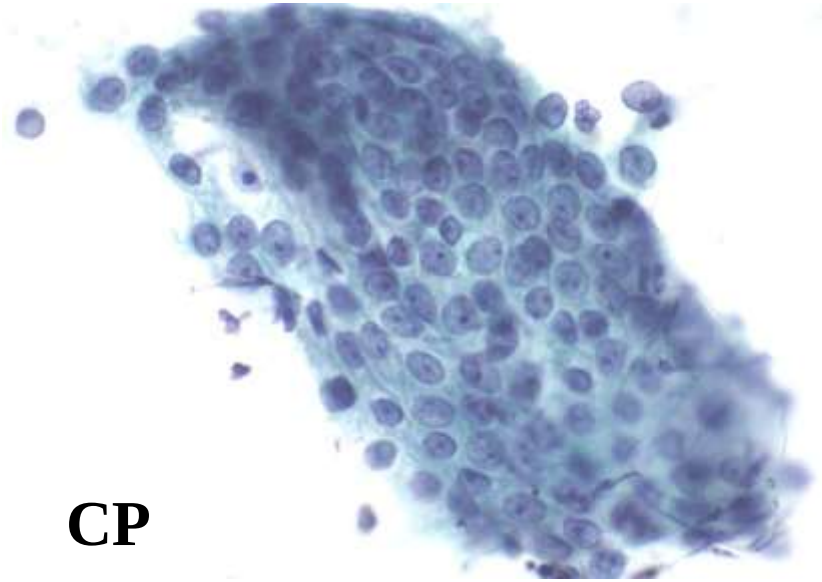




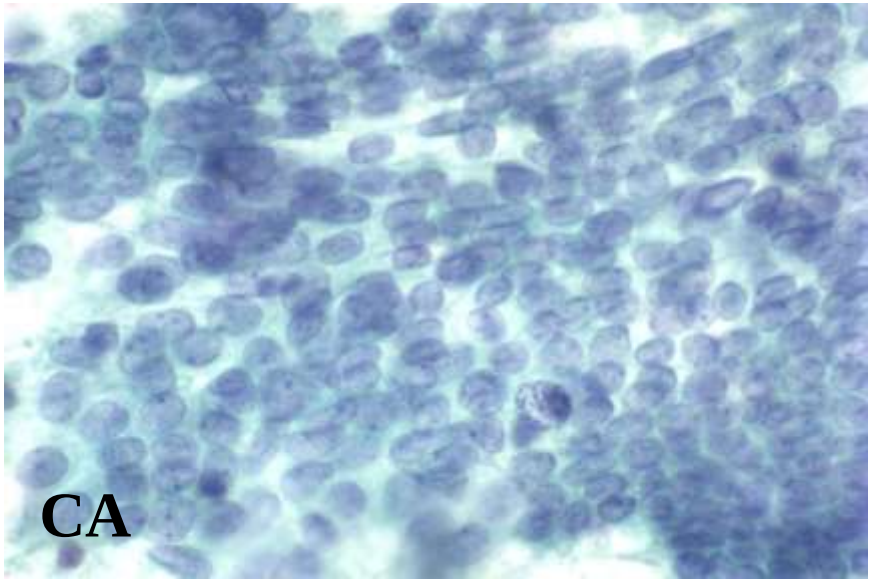


Chronic Pancreatitis

- mostly ductal cells
- scantily cellular
- some islet cells
- monolayered sheets
- cohesive, few single cells
- maintained polarity
- minimal nuclear overlap
- mild anisonucleosis
- smooth nuclear membranes
- rare/normal mitoses
- no coagulative necrosis



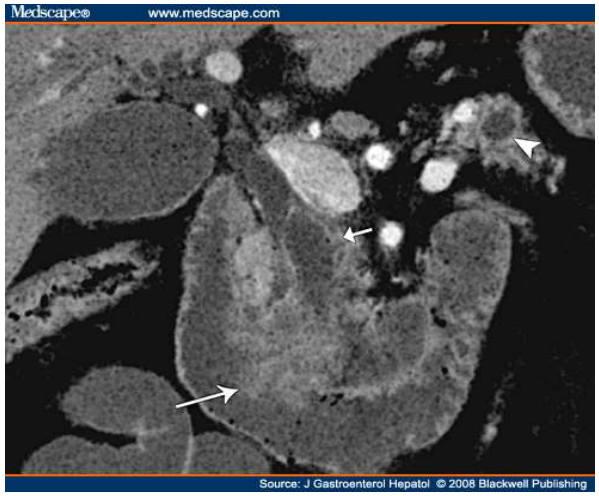
CP



CA

Clinical and Radiological Features of PDAC

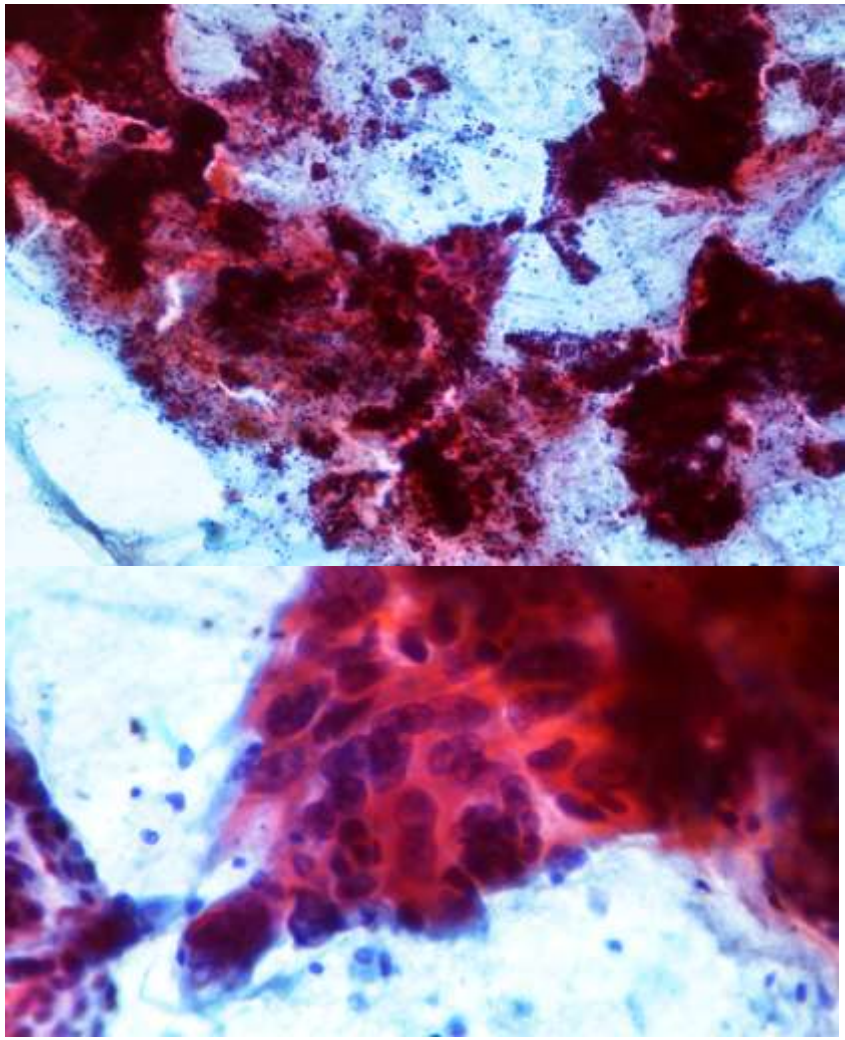
- 60-80 y.o. M>F
- Radiating epigastric pain with wt. loss
- Jaundice
- Migratory thrombophlebitis
- Sudden onset DM
- Double duct sign on CT
- Hypodense mass in panc head with irregular borders; atrophy elsewhere



- Cigarette smoking
- Long term DM
- Family history
- Germline mutations
  - PJS [*STK11/LKB1*]: 132x (Peutz-Jeghers syndrome)
  - FAMMM [*p16/CDKN2A*] (familial atypical multiple mole melanoma)
  - FANC
  - BRCA2
  - Familial CP [*PRSS1/SPINK1*]

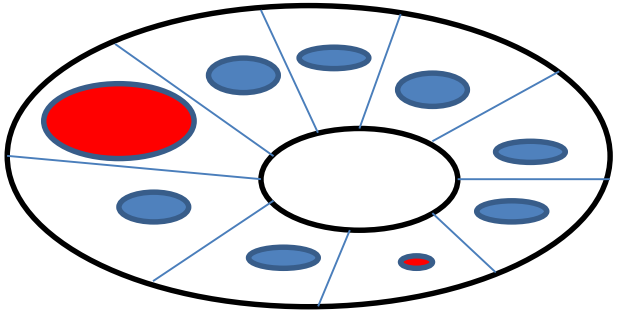
Pancreatic Ductal Adenocarcinoma
General Diagnostic approach

- Present as a solid mass
- 85 to 90% of all pancreatic cancer
- **Low power**
  - Cellularity
  - Cellular arrangement
  - Cohesiveness
  - Background
- **Intermediate power**
  - Composition of the cells groups
  - Organization of the cells groups
    - Polarity
    - Crowding
    - 2D VS 3D
- **High power**
  - Nuclear features
    - Size ▪ Contours ▪ N:C ratio
    - Chromatin ▪ Anisonucleosis ▪ Nucleoli ▪ Mitoses



Cytological features of PDC

- **High cellularity**
- **Background: clean, inflammatory mucinous and necrotic**
- **Predominantly ductal cells**
- **Cells groups with overcrowding and / or disorderly arrangement**
  - Large 2D → "drunken honeycomb → 3D groups → Aanisonucleosis >4:1 within single groups

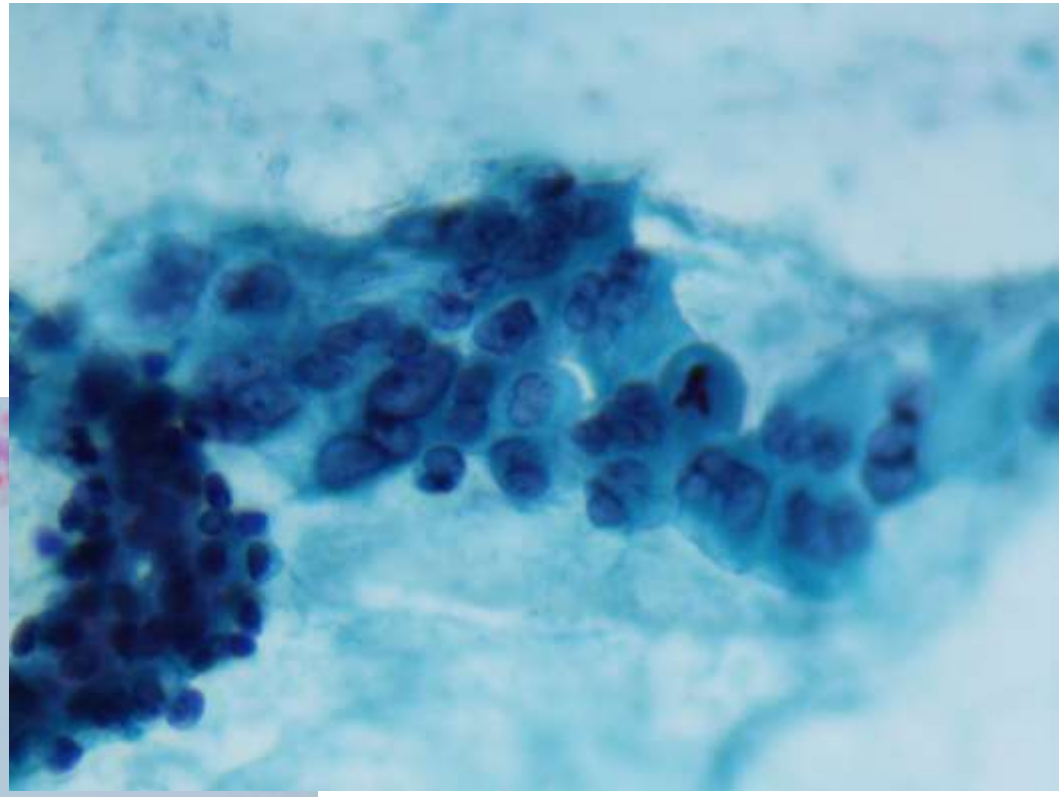


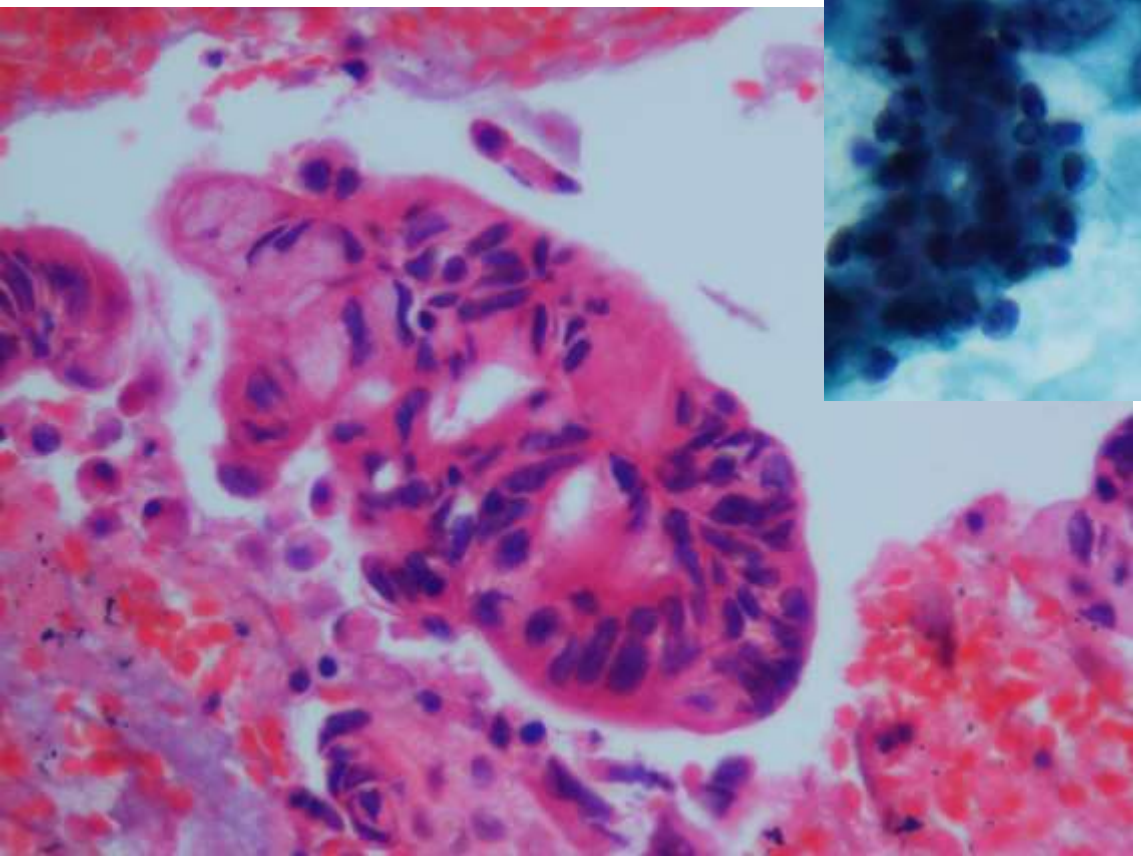
- **Isolated atypical cells**
- **Nuclear atypia**
  - Nuclear enlargement (>2x the size of a red blood cell)
  - Irregular contours
  - Coarse chromatin
  - Macronucleoli
  - Bi - and multinucleation
  - Mitotic figures

Immunohistochemistry

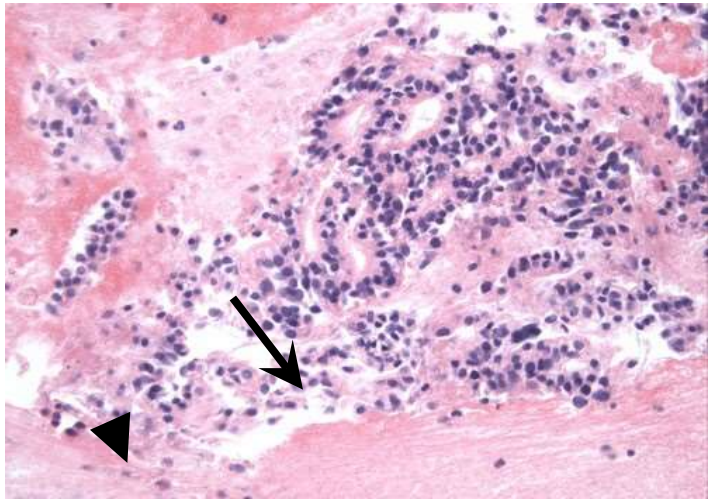
CEA (+)
Ca19-9 (+)
Ca125 (+)
CK7 (+)
CK19 (+)
Maspin (+)
Mucicarmine (+)
pVHL (-)
CK20 (-)

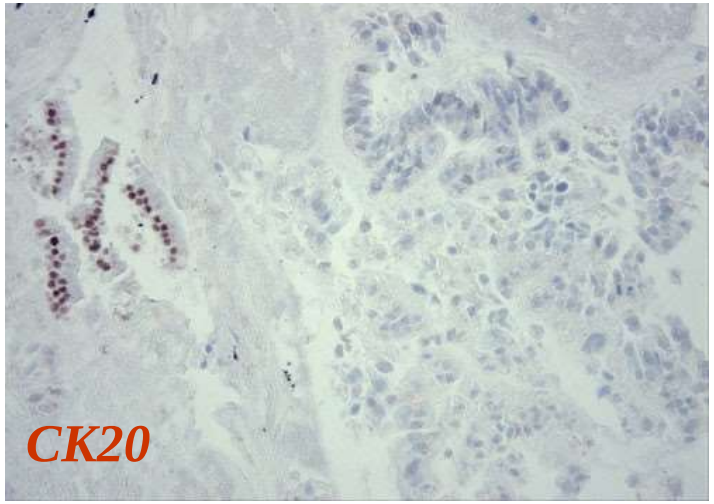
Pancreatic Ductal Adenocarcinoma

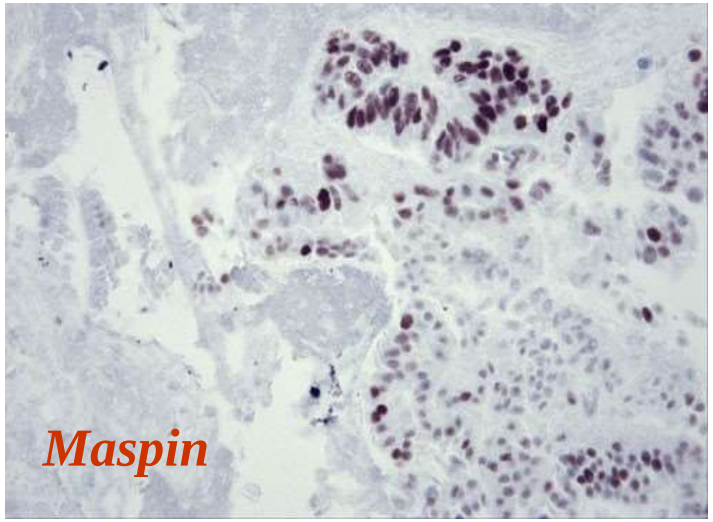


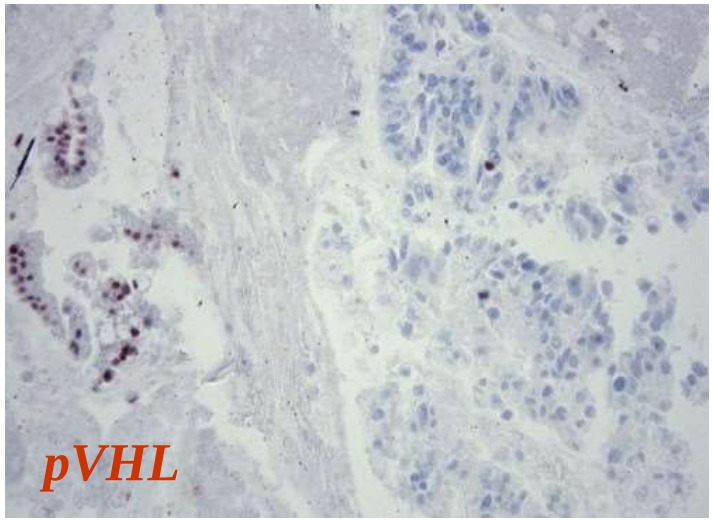


The Immunohistochemical Expression Pattern of Maspin, pVHL, and CDX2 is Helpful in Diagnosing Pancreatic Ductal Adenocarcinoma in Endoscopic Ultrasound-Guided Fine Needle Aspirations



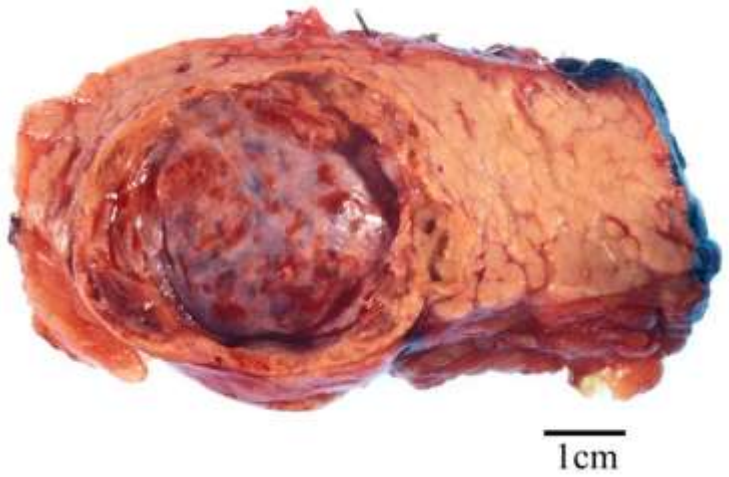


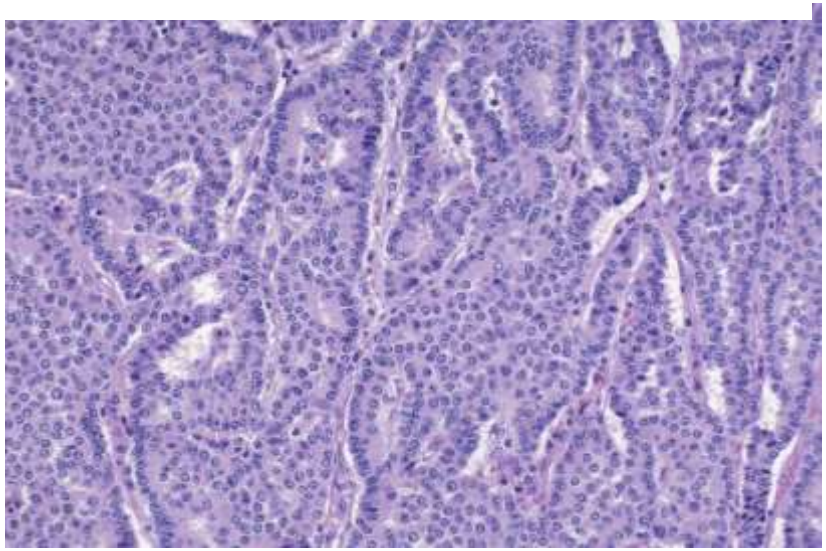




Pancreatic Neuroendocrine Tumor [PanNet, aka PEN, PET]

- **Clinical**
  - **Any age; 40-50 y.o.**
  - **M=F**
  - **MEN, VHL syndromes**
  - **Hormone effects in functional PanNet: insulin, glucagon for example**
- **Radiological**
  - **Pancreatic tail>>head/body**
  - **Round, well-circumscribed**
  - **Sometimes cystic, CA++**
  - **Octreotide scan+**
- **Histology**
  - **Cellular monomorphic population of polygonal cells with various organoid patterns with scan stroma (occasionally hylanized or amyloid stroma)**





Images: AFIP Pancreas fascicle 2007

Cytological Features of Pancreatic Endocrine tumours

<u>Typical Features</u>

Cellular aspirate

Loosely cohesive cell groups

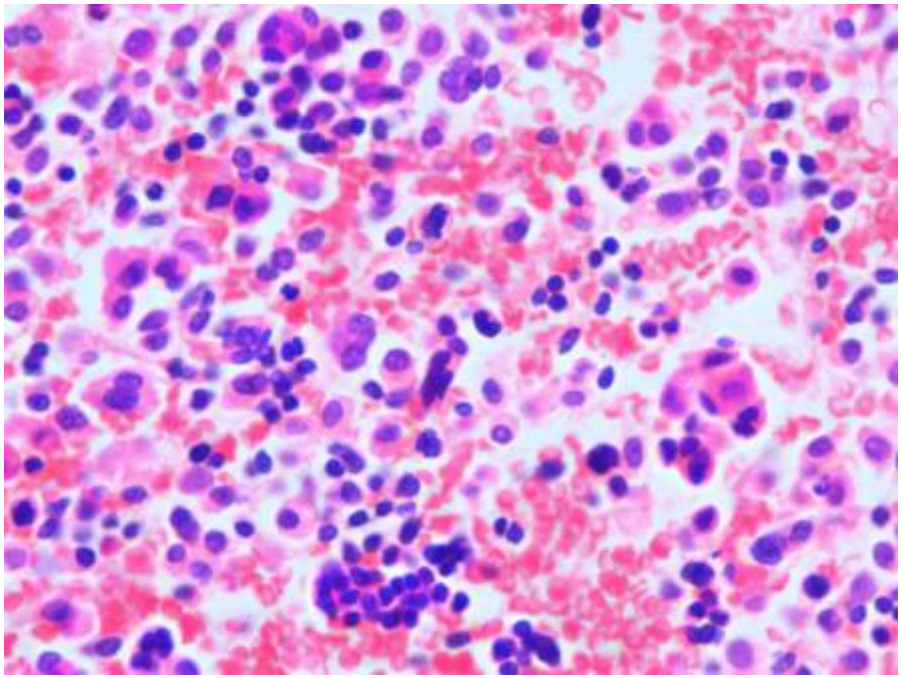
Rosette or pseudorosette formation

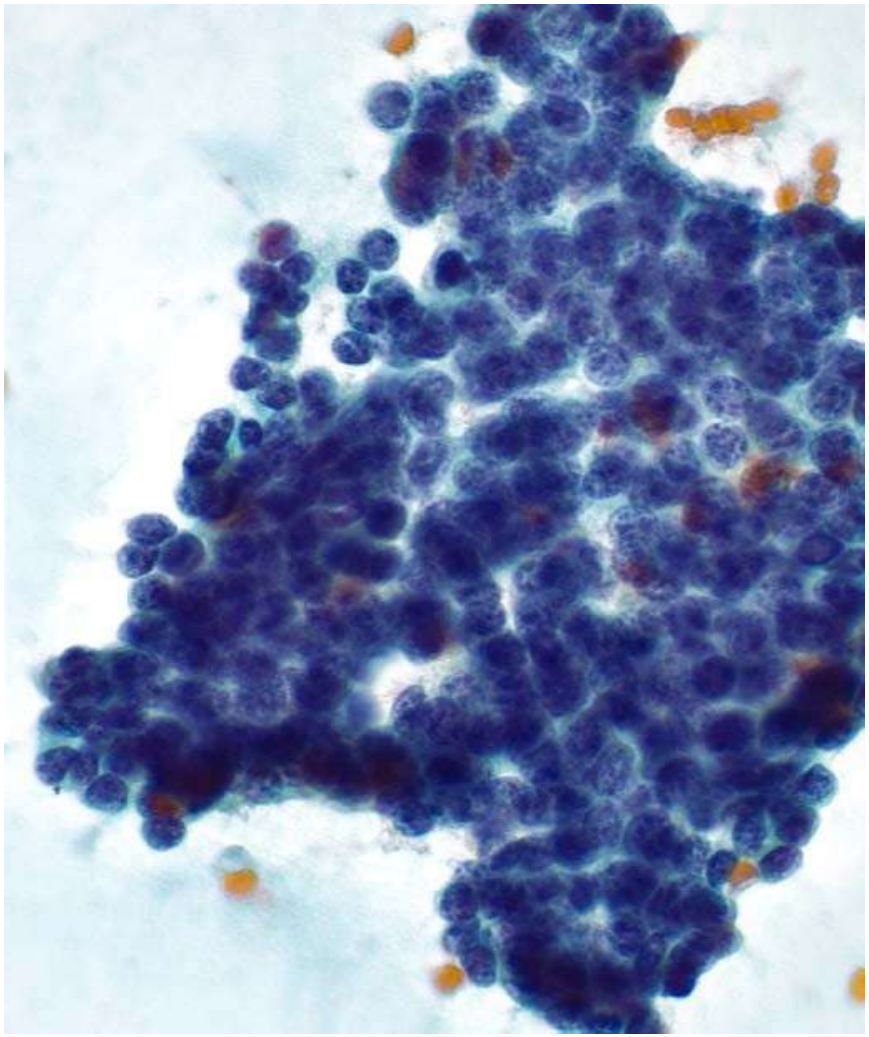
Relatively uniform, round-to-polygonal tumour cells

Plasmacytoid cells

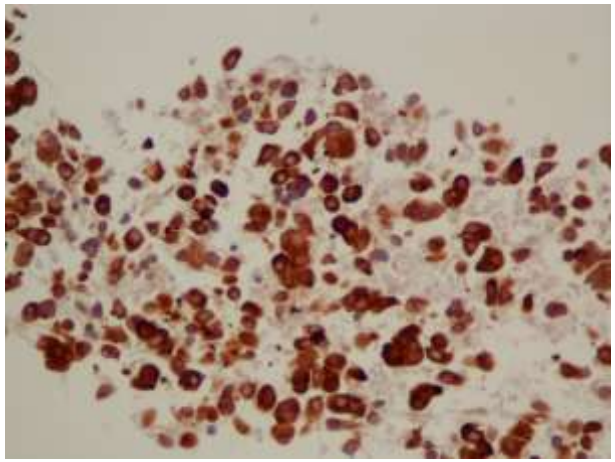
Salt-and-pepper chromatin

+/- nucleoli

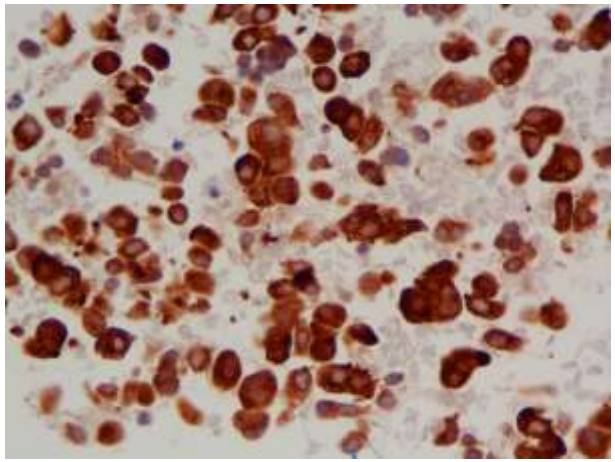




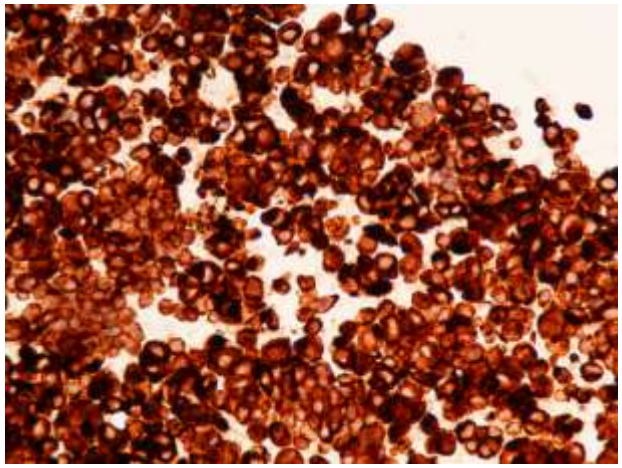
Pancreatic endocrine Tumour



Chromogranin



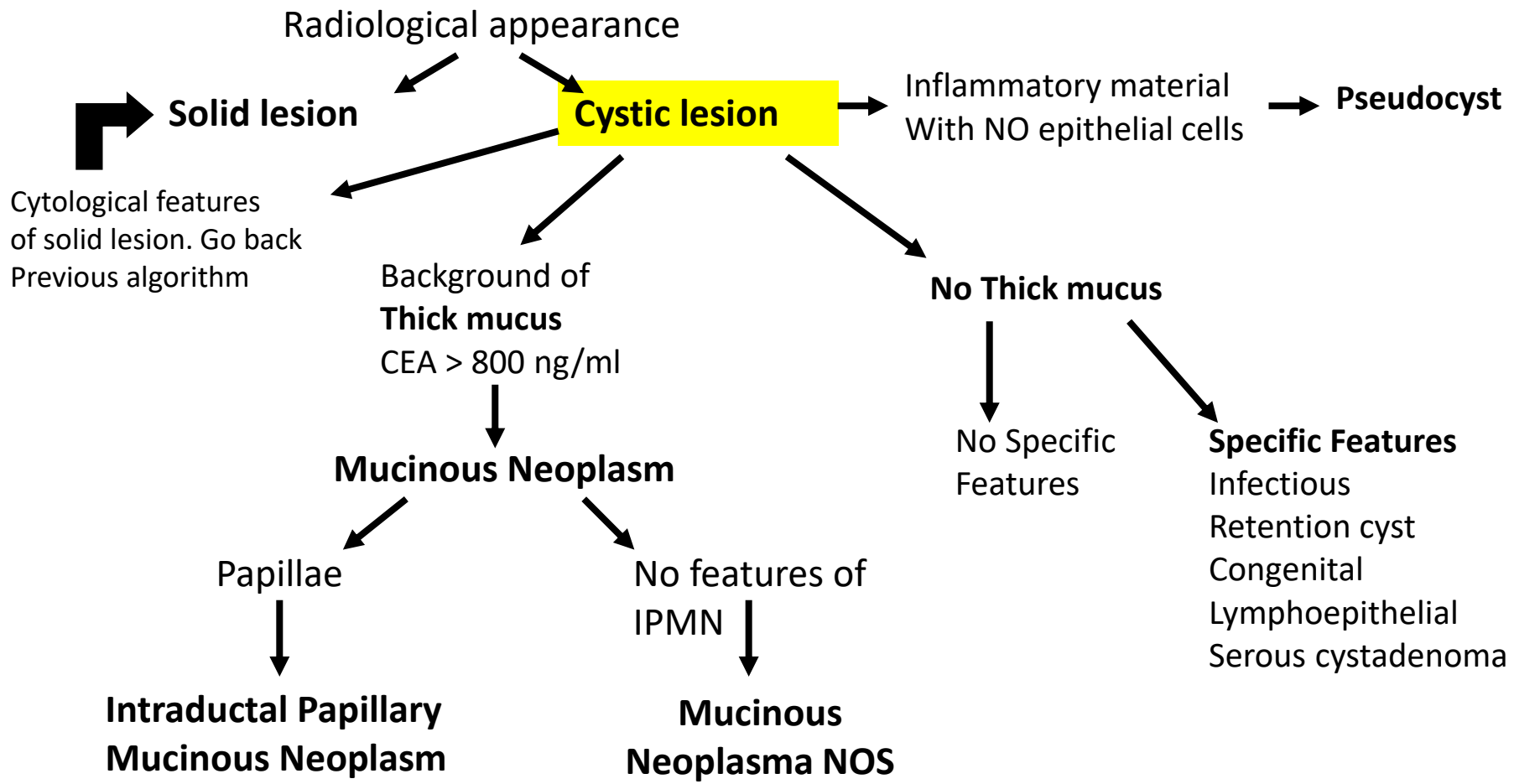
Synaptophysisn



Cam 5.2

	Neuroendocrine	*Pseudopapillary*
NSE	+	-
Chromogranin	+	-
Synaptophysisn	+	-
CD56	+	-
B-Cathenin	*Membrane +*	*Nuclear +*
CD10	_	+

Pancreatic Cytopathology. A Pragmatic Approach



Cysts of the Pancreas

Non-neoplastic

Pseudocyst
Abscess
Infectious
Retention
Lymphoepithelial cyst
Congenital

Neoplastic

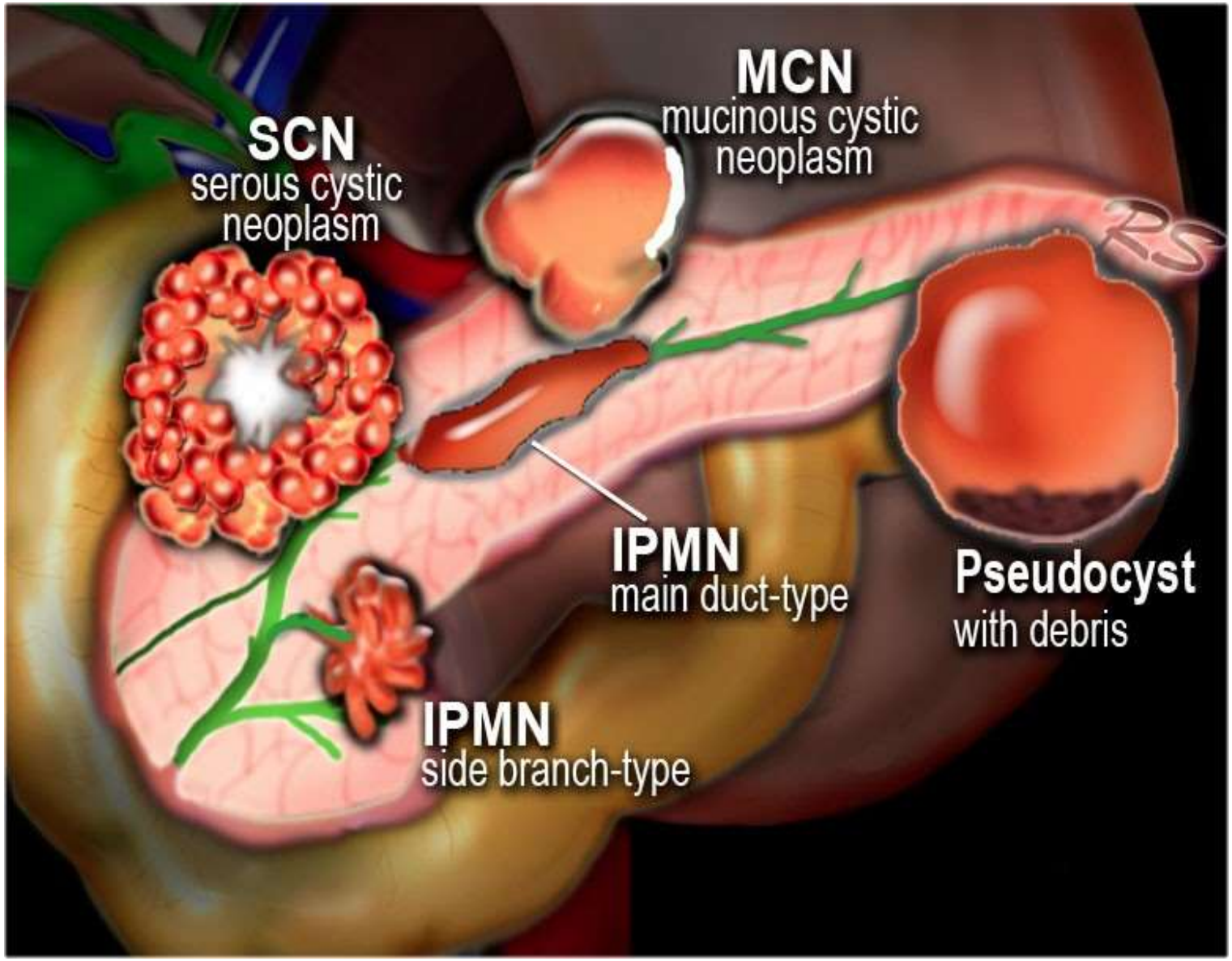
Mucinous cystic neoplasia
Intraductal papillary mucinous neoplaia
Serous cystic neoplasia
Benign vascular neoplasia
Typically solid neoplasia with cystic changes

Two basic questions for Cyst analysis

1) Is the cyst mucinous or non-mucinous?

2) Is the cyst low-grade or high-grade?

Pancreatic Cysts



Pancreatic Pseudocyst

Clinical

Most common cystic lesion in the pancreas

Associated with pancreatitis, trauma, surgery (almost always)

Radiology

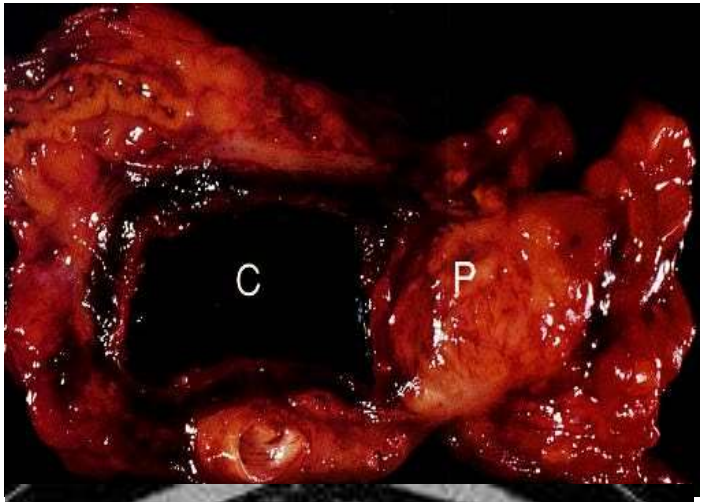
Unilocular, non-septated

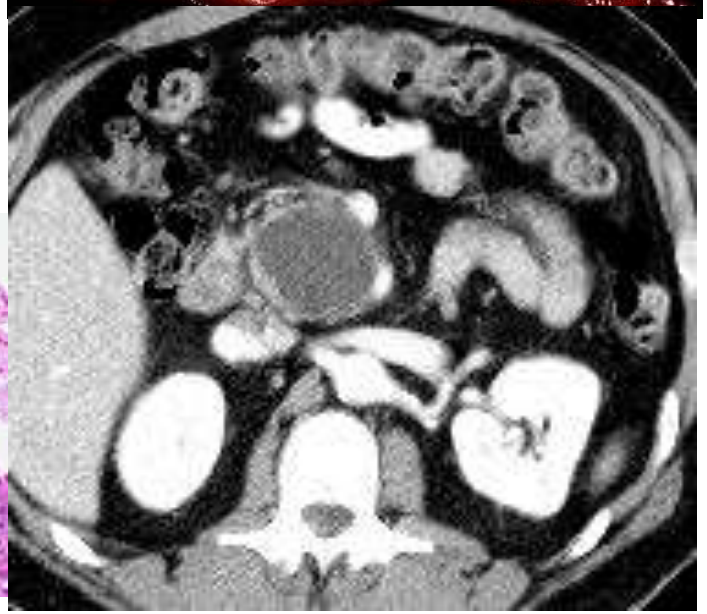
Thick to thin walled

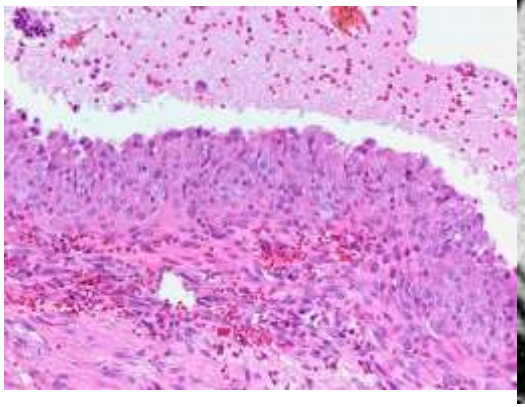
No mural nodule

Histology

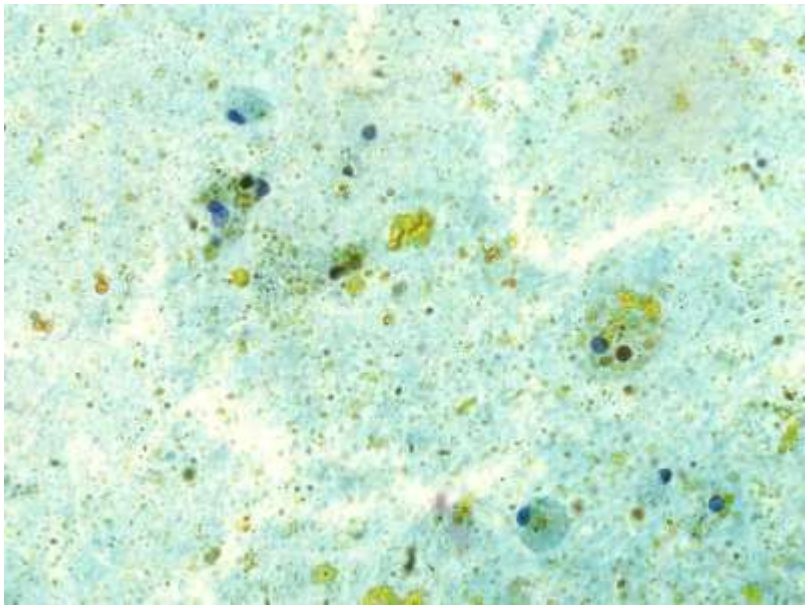
Cyst lining of histiocytes and inflammatory cells

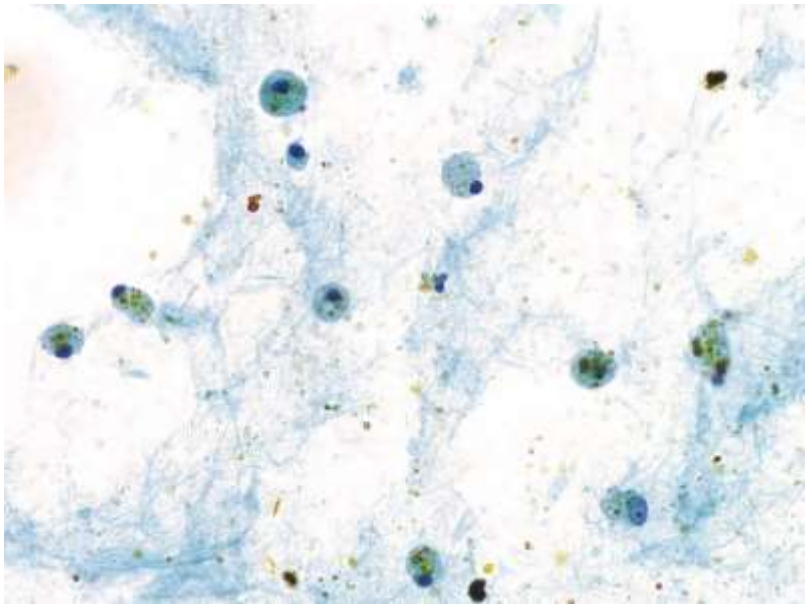






Pancreatic Pseudocyst
cytology





•cyst debris with blood, proteinaceous material and yellow hematoidin-like pigment (grossly brown and thin fluid)

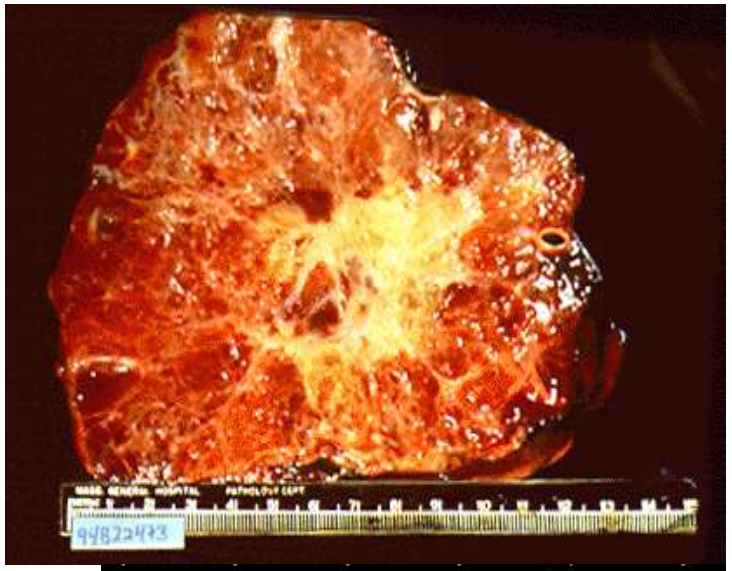
• variable inflammation

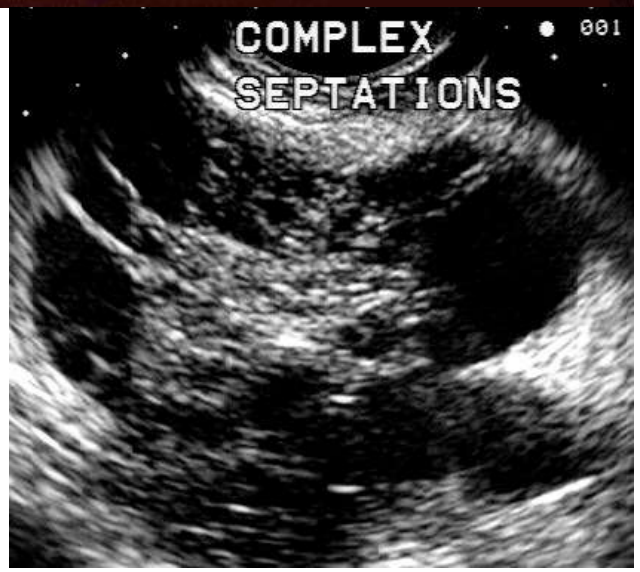
•NO cyst lining epithelium (beware of contamination, mucin and epithelium)

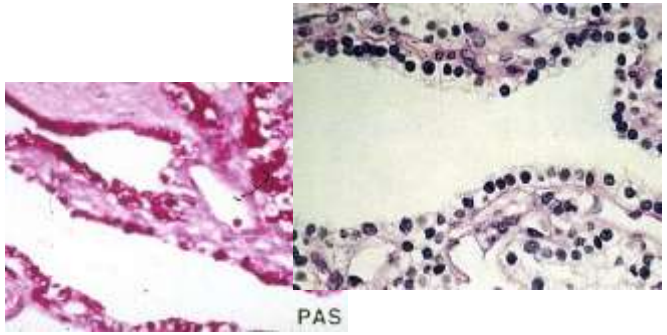
•CEA low; amylase usually in the 1000's; no *KRAS*

Serous Cystadenoma

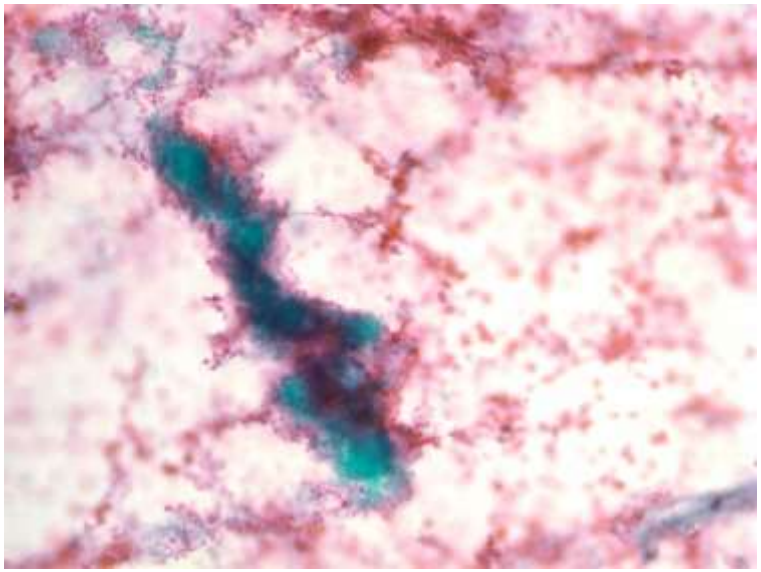
- **Clinical**
  - **Benign, slow growing neoplasm women>>men, mean age 7th decade**
  - **Assocaited with VHL with deletion of 3p25 in most cases**
  - **Often asymptomatic, but can hemorrhage and cause pain**
- **Radiology**
  - **circumscribed, multi-lobulated**
    - **Microcystic with fibrous septae, central scar, calcifications in ~30-40%**
- **Histology**
  - **"glycogen-rich"**
  - **dPAS+ cuboidal**
  - **epithelium**

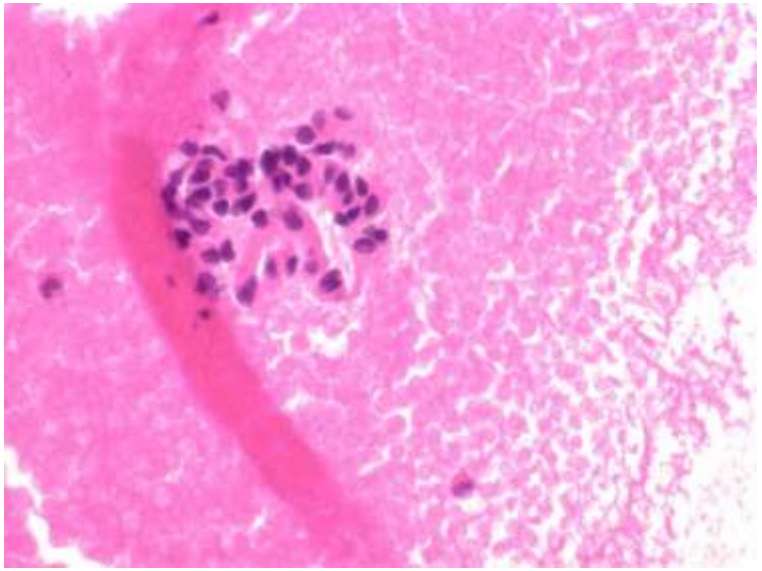


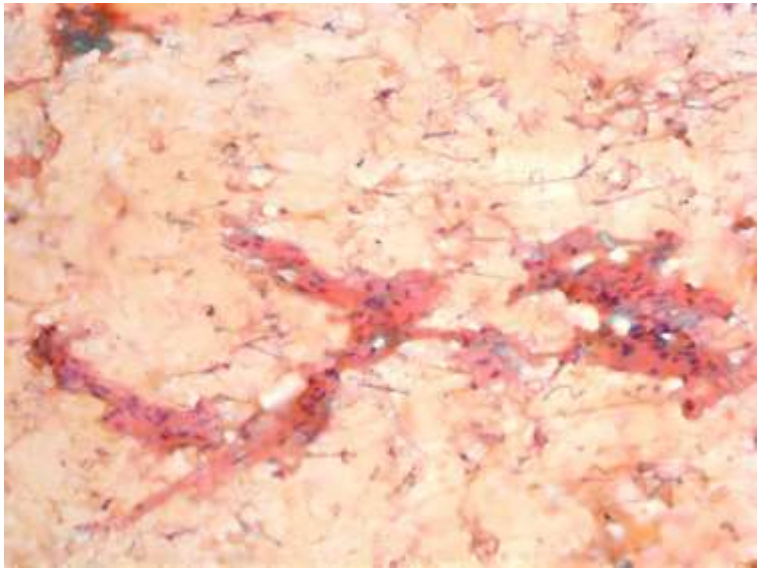


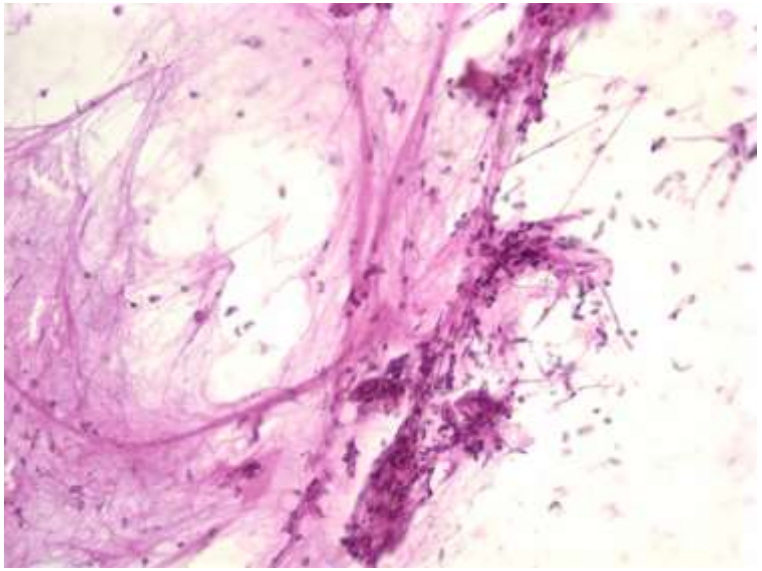


SCA



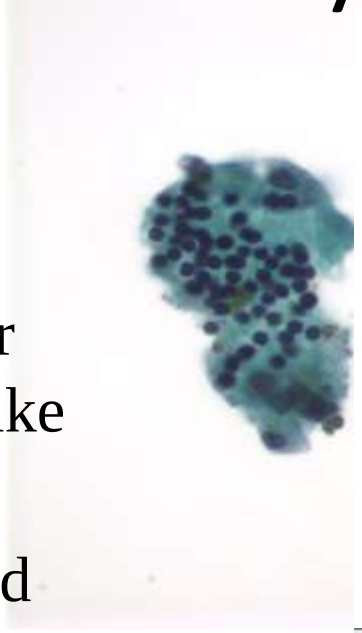


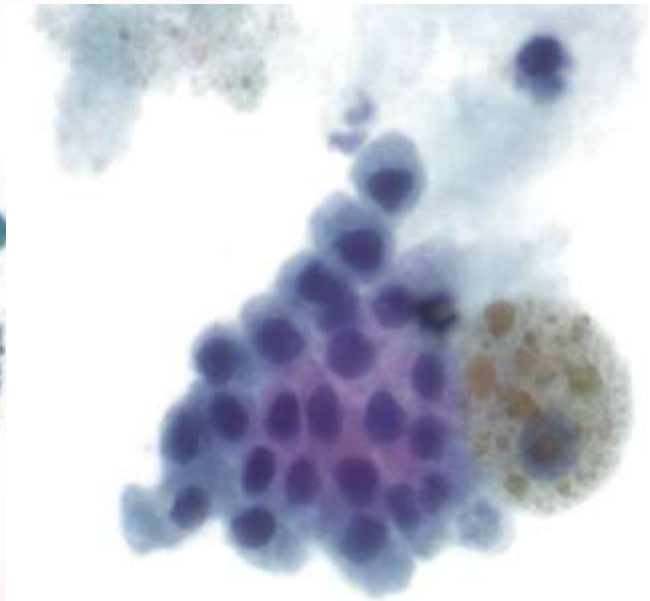


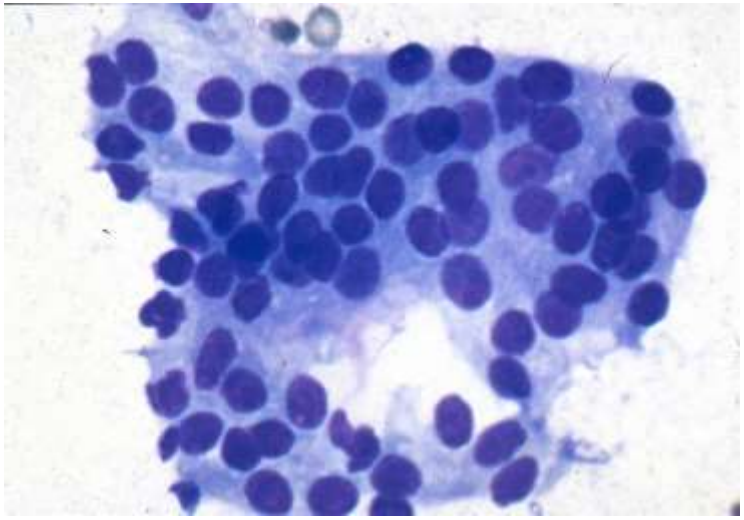


Serous Cystadenoma

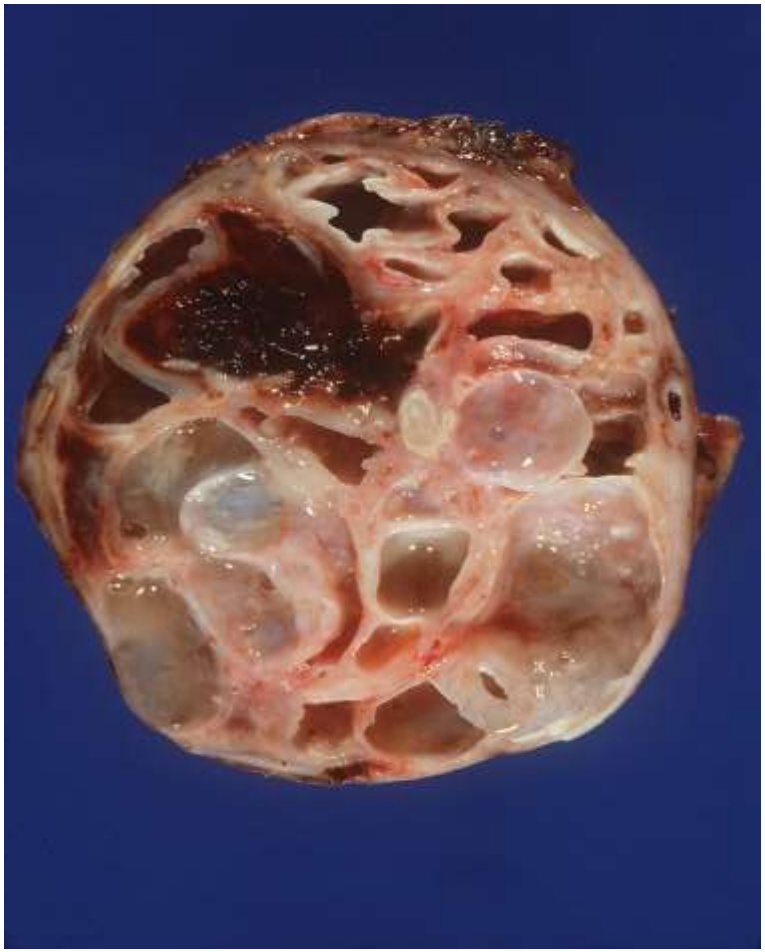
- Cuboidal non-mucinous epithelial cells
- Hemosiderin-laden macrophages in a clean or bloody, non-pseudocyst like background
- Grossly bloody or thin and clear
- CEA and amylase low
- NO *KRAS/GNAS*
- 3p deletions (3p25, VHL)



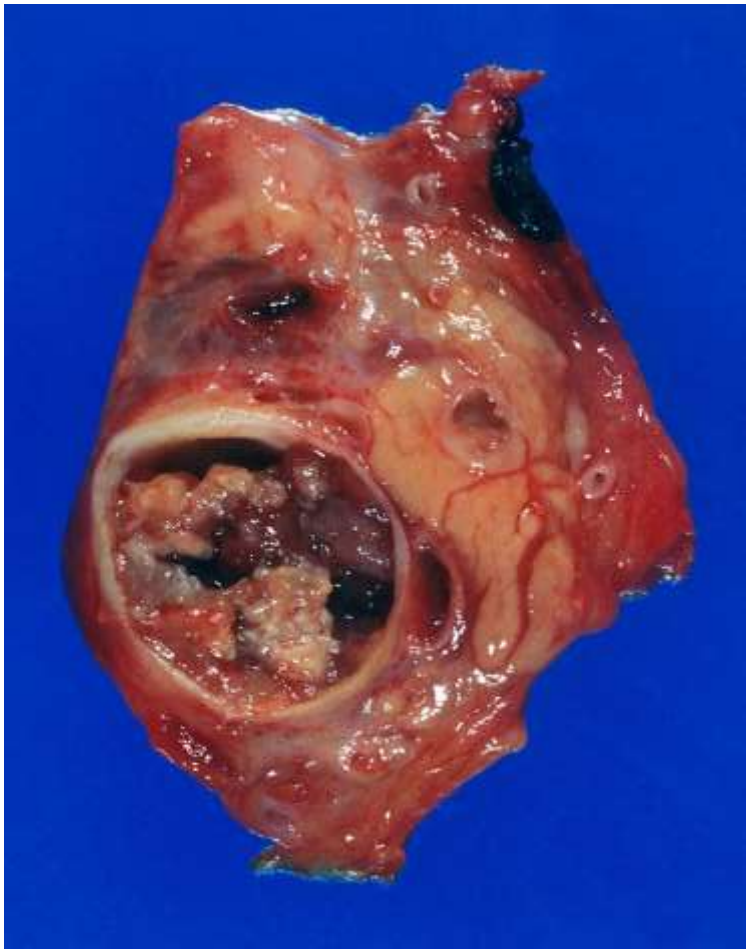




Neoplastic Mucinous Cysts



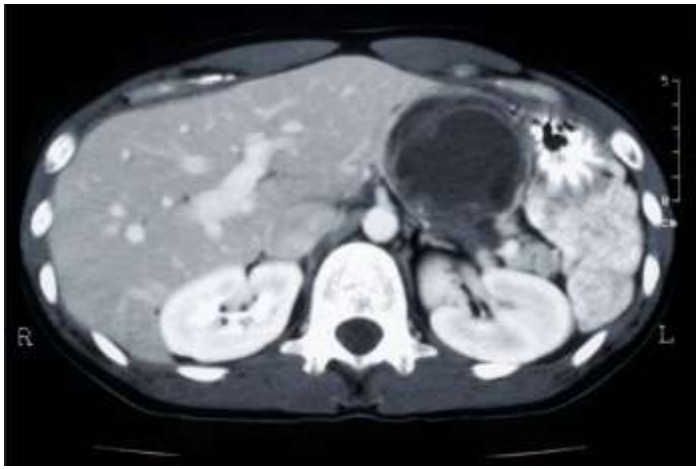
MCN

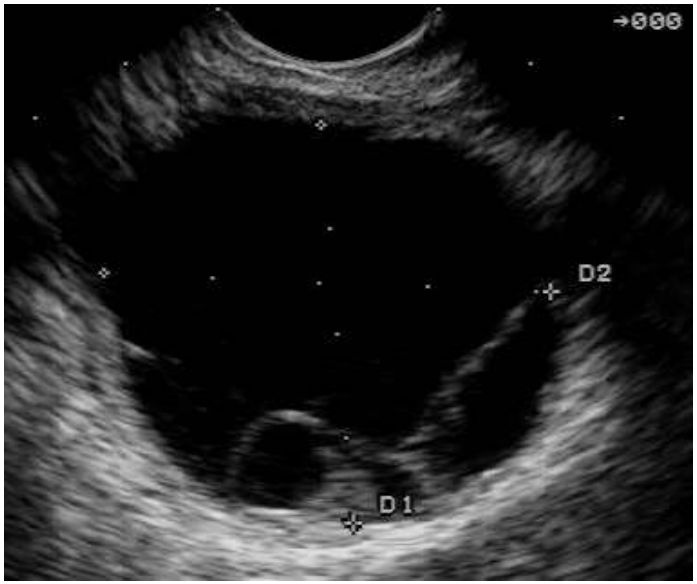


IPMN

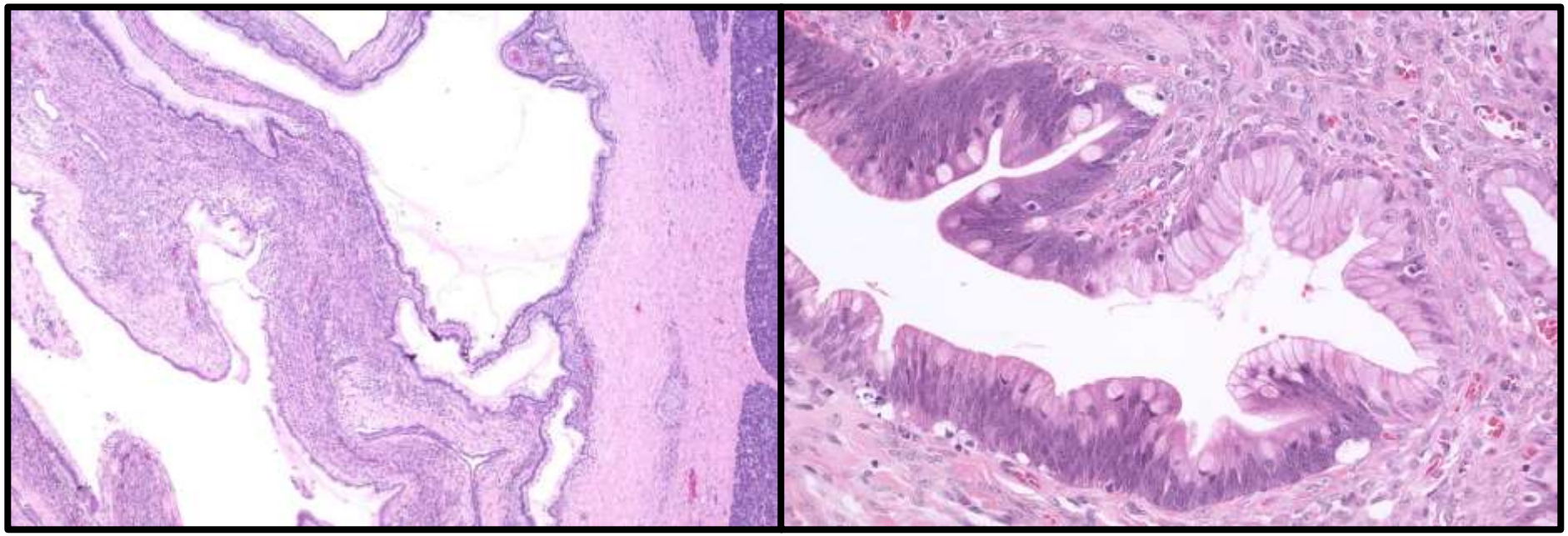
Mucinous Cystic Neoplasm

- Clinical
  - F:M=20:1
  - Most are benign
  - Prognosis excellent for non-invasive completely resected tumors
  - Resection recommended despite grade
- Radiology
  - body and tail (90%)
  - do not communicate with the pancreatic ductal system
  - thick walled (Ca++ in 20%)
  - thin or thick septa





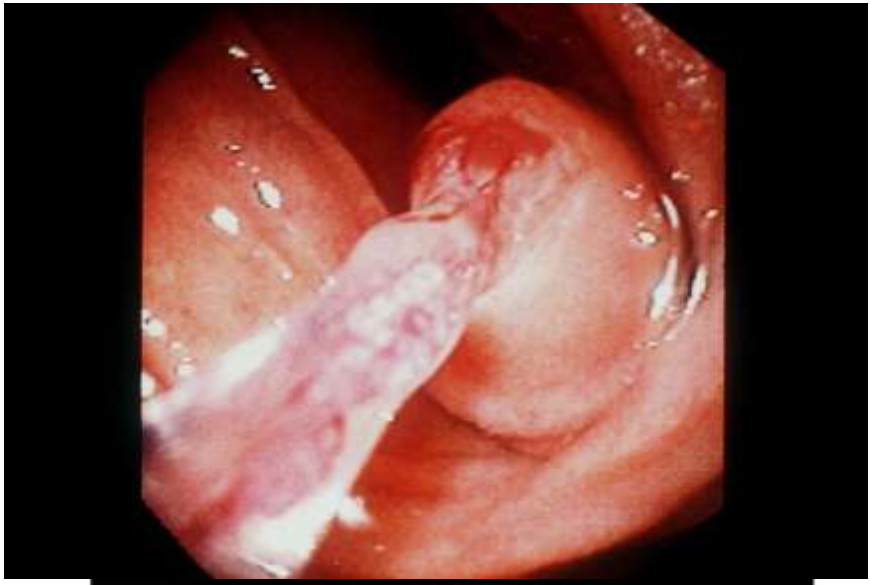
Mucinous Cystic Neoplasm

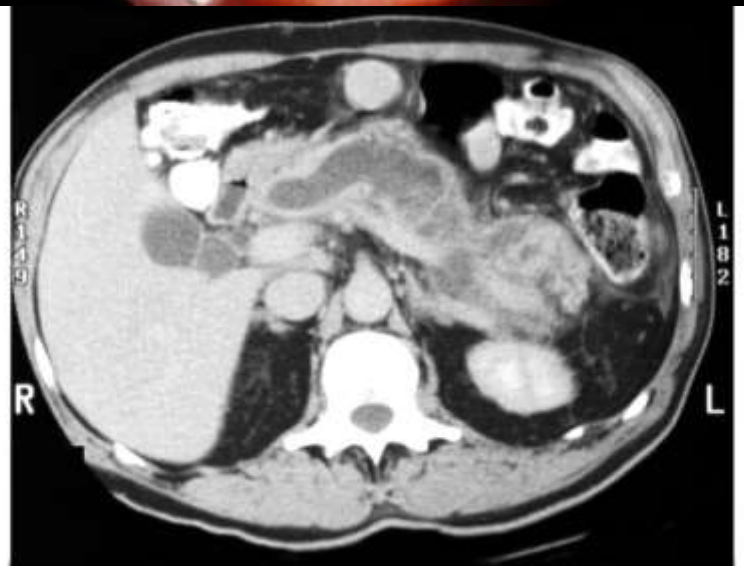


- Not associated with the pancreatic ducts
- Lined by mucinous, generally non-papillary epithelium
- Subepithelial "ovarian-like stroma" <u>required</u>
- Atypia may be very heterogeneous; invasion may be very focal, so the entire cyst should be submitted for histology

IPMN

- Main duct type
  - Diagnosed clinically
  - Dilated main pancreatic duct (definition varies, but >5mm)
  - Pancreatic head mostly, but occur all through the pancreas
  - Intestinal type lining most common
  - 60% have HGD
  - 45% have invasive carcinoma
  - Symptoms common but 25% asymptomatic
  - Treatment-resection

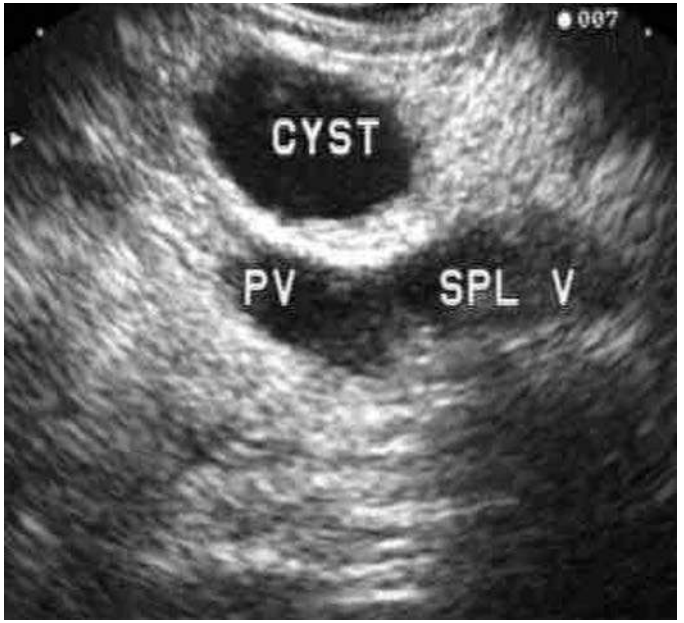


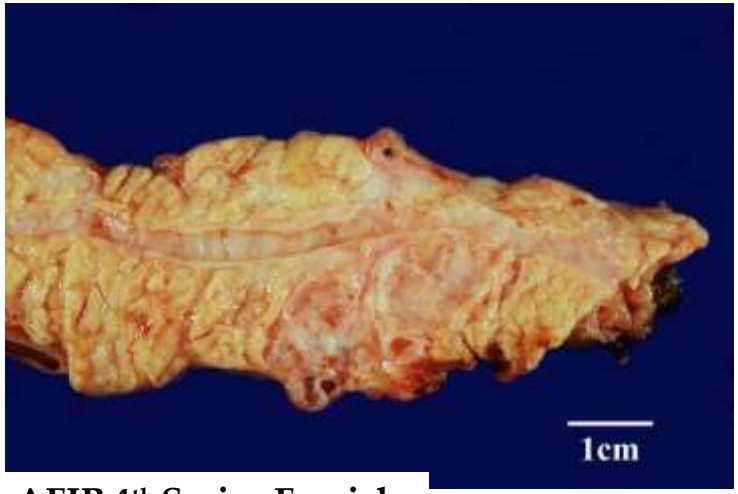


AFIP 4th Series Fascicle

IPMN

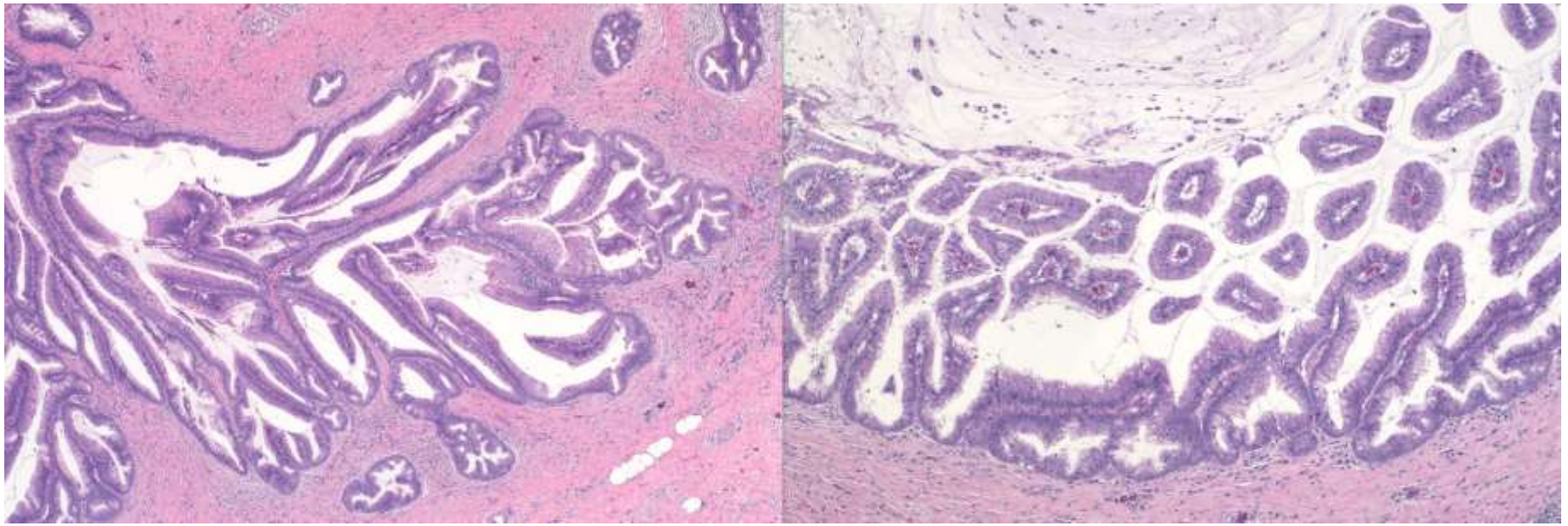
- Branch Duct Type
  - Most often in head/uncinate
  - 1/3 with multiple cysts
    - Supports clinical dx
  - Most patients asymptomatic
  - Imaging: “bunch of grapes”; single cyst may not be diagnostic for BD-IPMN unless visualized connection to the MPD
  - Most lined by gastric type epithelium
  - Most low grade
  - Treatment-depends….





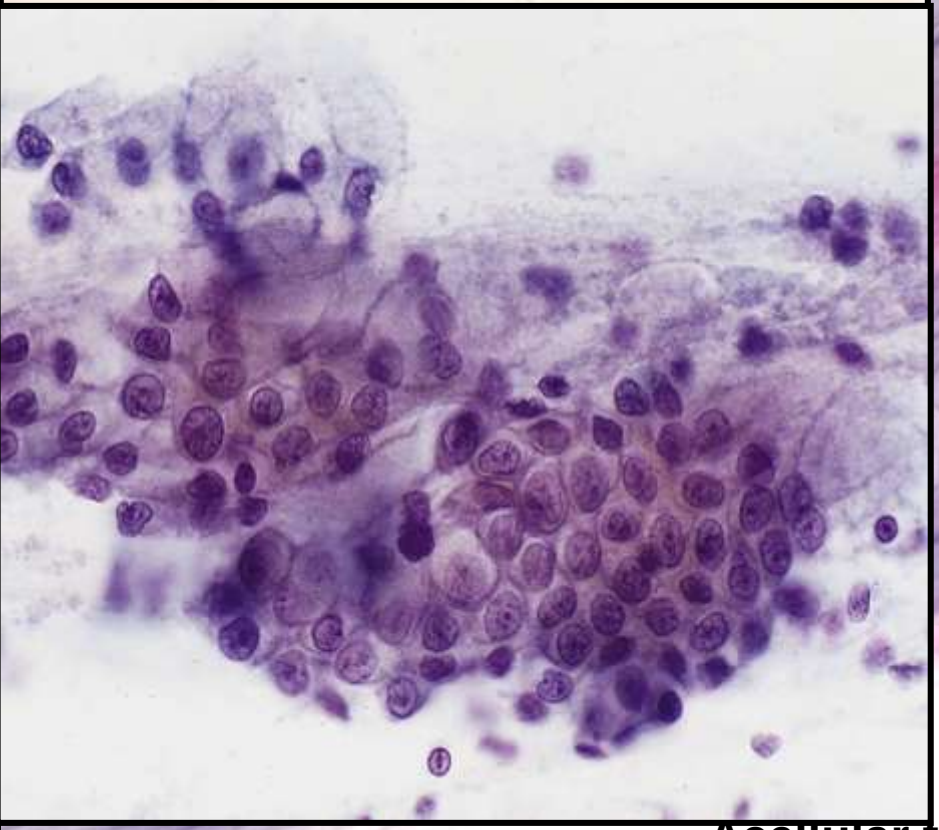
AFIP 4th Series Fascicle

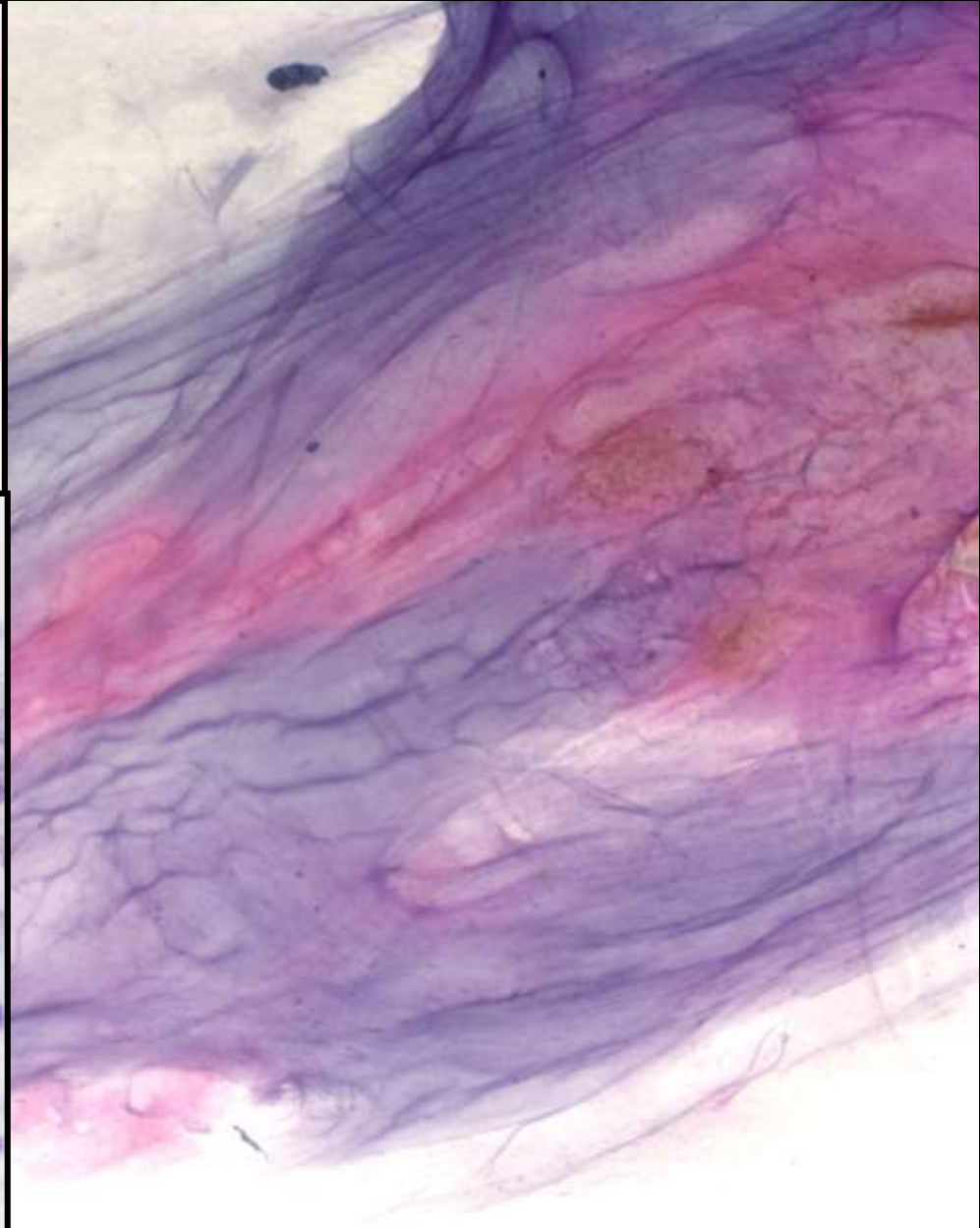
IPMN



- Variously papillary mucinous epithelium of variable cell type and heterogenous atypia
- No association with ovarian-like stroma under the epithelium

AFIP 4th Series Fascicle





Acellular thick, colloid-like mucin is NOT non-diagnostic!

BIOCHEMICAL AND MOLECULAR TESTS FOR CLASSIFYING A PANCREATIC CYST

Cyst	CEA	Amylase	*KRAS*	*GNAS*
Pseudocyst	↓	↑ ↑	-	-
Serous cystadenoma	↓	↓	-	-
Intraductal papillary mucinous neoplasm	↑	↑	+	+
Mucinous cystic neoplasm	↑	↑ ↓	+	-

Questions to answer before you report your pancreatic cytology

It is solid of cystic?

It is a man or a women?

Age?

Location

a) **Head?**
b) **Neck?**
c) **Body?**

What sort of sampling?

a) **Brushings?**
b) **FNA?**

Any relevant history?

a) **Alcohol abuse?**
b) **PSC?**
c) **Autoimmune pancreatitis?**
d) **Stents?**
e) **Stones?**

STANDARDIZED NOMENCLATURE FOR PANCREATIC CYTOLOGY PROPOSED BY THE PAPANICOLAOU SOCIETY OF CYTOPATHOLOGY

I. NONDIAGNOSTIC

II. NEGATIVE

- Pancreatitis
- Lymphoepithelial cyst
- Splenule/accessory spleen
- Pseudocyst
- Benign pancreatic tissue with no discrete mass lesion

I. ATYPICAL

II. NEOPLASTIC

- Benign
- Serous cystadenoma
- Other
- Pre-malignant mucinous cysts (MCN and IPMN) – specify low- or high-grade cellular atypia
- Well-differentiated neuroendocrine tumors
- Solid-pseudopapillary neoplasm

V. SUSPICIOUS

VI. POSITIVE/MALIGNANT

- Ductal adenocarcinoma and variants
- Acinar cell carcinoma
- Pancreatoblastoma
- Lymphoma
- Metastatic