Biliary cytology and pancreatic endoscopic ultrasound-guided FNA

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Reasons for biliary cytology

- PSC- is a pre-neoplastic condition in young individuals, the cure of which is liver transplantation before the development of cancer. One of the leading causes for liver transplantation in the Nordic countries.

- Other causes for malignancies in the gall-ways are
  - Congenital malformations
  - Papillomatosis
  - Adenomas
  - Parasites
  - Stones
The transplantation must take place before malignant transformation

• We have to catch the disease when still "in situ"

• In theory this is possible by fiberoptics; We have to learn to recognise dysplasia
Primary sclerosing cholangitis (PSC)
PSC

• Progressive disease - median survival from diagnosis to death is about 18 years (Björö J Hepatol 2004)

• Liver transplantation can cure the disease

• Cholangioca evolves in 6-18% 

• Cholangioca is kontraindikation for liver transplantation

=> the patients, who will get cholangioca, should be found in time
PSC and IBD

• 80-90% of those with PSC have IBD also

• Also the risk of colonic cancer is increased in patients with IBD and PSC:
  - 10 years 9%, 20 years 31% and 25 years 50% (Broome, Hepatology 1995, Gurbuz 1995, Loftus 1996, Nuako 1998)
  - 10 years 14% and 20 years 31% (Claessen, J Hepatol 2008)

⇒ Yearly colposcopies are mandatory
Additional value of DNA-ploidy in evaluation of malignant and premalignant biliary strictures in patients with PSC

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Leena Krogerus, pathology
Kirsti Numminen, radiology
Heikki Mäkinsalo, surgery
Martti Färkkilä, gastroenterology

Helsinki University Hospital
Patients (n=103):

Consecutive PSC patients with extrahepatic strictures or clinical suspicion of biliary malignancy.

End point (hepatobiliary malignancy or examination of explanted liver) achieved in 40 (39%) patients.
ERC-score
(Ponsioen et al. Gut 2002;51:562-566)

Intrahepatic
0  No visible abnormalities
I   Multiple strictures; normal calibre of bile ducts or minimal dilatation
II  Multiple strictures, saccular dilatations, Decreased arborisation
III Only central branches filled despite adequate filling pressure; severe pruning

Extrahepatic
0  No visible abnormalities
I   Slight irregularities of duct contour, no stricture
II  Segmental stricture
III Stricture of almost entire length of duct
IV  Extremely irregular margin; diverticulum-like out-pouchings
DNA PLOIDITY ANALYSIS

Aneuploid DNA content: 20 patients
- 12 patients: Hepatobiliary malignancy or dysplasia in the bile ducts
- 1 patient: No dysplasia
- 7 patients: In follow up

Diploid DNA content: 83 patients
- 12 patients: Hepatobiliary malignancy or dysplasia in the bile ducts
- 15 patients: No dysplasia
- 56 patients: In follow up
Can DNA cytometry be used for evaluation of malignancy and premalignancy in bile duct strictures in primary sclerosing cholangitis?

Bergqvist et al. J Hepatol 2000;33:873-877

• **Benign bile ducts vs CC (PSC and non-PSC):**
  12% (2/17) vs 54% (15/28), p < 0.005

• **Benign bile ducts vs CC in PSC:**
  12% (2/17) vs 80% (8/10), p < 0.0007
Lindberg et. al. Endoscopy 2002;34:909-916

Patients:  - 25 benign strictures (13 PSC)
          - 32 malignant strictures (7 PSC+CCA)

<table>
<thead>
<tr>
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<th>Sensitivity</th>
<th>Specificity</th>
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<tbody>
<tr>
<td>Cytology</td>
<td>55 %</td>
<td>100 %</td>
</tr>
<tr>
<td>DNA analysis</td>
<td>52 %</td>
<td>96 %</td>
</tr>
<tr>
<td>Ca 19-9</td>
<td>67 %</td>
<td>89 %</td>
</tr>
<tr>
<td>CEA</td>
<td>56 %</td>
<td>89 %</td>
</tr>
<tr>
<td>Combination</td>
<td>88 %</td>
<td>80 %</td>
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Lindberg et. al. Endoscopy 2006; 38: 561-5

Patients: - 159 patients with biliary strictures

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<thead>
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<th>Test</th>
<th>Sensitivity</th>
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<tbody>
<tr>
<td>DNA analysis</td>
<td>43%</td>
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<tr>
<td>Cytology</td>
<td>57%</td>
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<tr>
<td>DNA analysis + cytology</td>
<td>62%</td>
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Conclusion: DNA aneuploidy is a marker of poor prognosis in patients with malignant biliary strictures
Re-examination of brush cytology from the biliary tree
(Re-examined by JA and LK, κ 81%)

Malignant cells: no patients

Cells suspected for malignancy: 21 patients
- 11 patients: Hepatobiliary malignancy or dysplasia in the bile ducts
- 2 patients: No dysplasia
- 8 patients: In follow up

Benign cells: 82 patients
- 13 patients: Hepatobiliary malignancy or dysplasia in the bile ducts
- 14 patients: No dysplasia
- 55 patients: In follow up
The end-point achieved (40 patients):
- 24 hepatobiliary malignancy or dysplasia
- 16 no dysplasia

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<th>Test</th>
<th>Sensitivity</th>
<th>Specificity</th>
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</thead>
<tbody>
<tr>
<td>Cytology</td>
<td>48%</td>
<td>87%</td>
</tr>
<tr>
<td>DNA analysis</td>
<td>56%</td>
<td>93%</td>
</tr>
<tr>
<td>Combination</td>
<td>75%</td>
<td>81%</td>
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PSC and IBD

- IBD diagnosed in 74 (72%) of the 103 PSC patients
- Colorectal dysplasia or carcinoma in 16 (22%) of 74 IBD patients
- End point achieved in 8 patients with colorectal dysplasia/carcinoma: 4 (50%) hepatobiliary dysplasia/carcinoma found
Histology from choledochus with dysplasia
Potilas 2

Lymph nodes, removed by laparoscopy
Normal epithelium from the biliary tree, pap-stain on sytotechx10
Normal epithelium in a cellblock
Normal epithelium im MGGx40
Normal epithelium in a cellblock, H&Ex100
Normal, papx100
File analyzed: FC04014.001
Date analyzed: 4-Mar-2004
Model: 2D2A2n_DSD_ASD
Analysis type: Manual analysis

Diploid: 83.69 %
Dip G1: 86.06 % at 56.25
Dip G2: 5.97 % at 109.69
Dip S: 7.97 % G2/G1: 1.95
%CV: 5.29

Aneuploid 1: 16.31 %
An1 G1: 58.71 % at 99.39
An1 G2: 6.79 % at 193.81
An1 S: 34.50 % G2/G1: 1.95
%CV: 4.28 DI: 1.77

Total Aneuploid S-Phase: 34.50 %
Total S-Phase: 12.30 %
Total B.A.D.: 9.37 %

Stnd 1: 23.76 % at 19.60 DI: 0.35
Dip G1 Ratio: 2.87 %CV: 2.49
Stnd 2: 12.65 % at 41.15 DI: 0.73
Dip G1 Ratio: 1.37 %CV: 2.09

Debris: 16.71 %
Aggregates: 0.00 %
Modeled events: 18867
All cycle events: 9992
Cycle events per channel: 72
RCS: 6.645
Abnormal cell-group in a papx10
Brush, cytospin, pap x 20
Brush, cytospin, pap x 40
Brush, cytospin, pap x40
Aneuploid cell group, cytospin, MGGx40
Histology: Low grade dysplasia in adenoma, H&E x20
High grade dysplasia, H&Ex100
High grade dysplasia in adenoma

H&E x10
The difficulty is inflammatory atypia
Coresponding histology, H&Ex20
Endoscopic ultrasound guided FNA from the pancreas

K. Yamao et al.: EUS-FNAB: past, present, future
# Pancreatic tumors

<table>
<thead>
<tr>
<th>Benign</th>
<th>pre-malignant</th>
<th>Malignant</th>
</tr>
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<tbody>
<tr>
<td>Pseudocysts</td>
<td>Intraductal papillary and mucinous cyst</td>
<td>Pancreatic ductal ca.</td>
</tr>
<tr>
<td>Solid pseudopapillary n. Pancreatic endocrine n.</td>
<td>Foamy gland ca.</td>
<td></td>
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<tr>
<td>Autoimmune pancreatitis</td>
<td>Neuroendocrine ca.</td>
<td></td>
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<tr>
<td>Serous cystadenoma</td>
<td>Undifferentiated ca.</td>
<td></td>
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<tr>
<td>Retention cysts</td>
<td>Acinar cell ca.</td>
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<tr>
<td>Dermoid cysts</td>
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Typical examples of a clearly malignant (A) and a nondiagnostic (B) finding illustrating the limitations of cytologic analyses. Both samples were obtained by FNAB of suspect pancreatic masses and analyzed within the course of this study. A, sample from “Biopsy___3” (see Table 1): malignant cells (arrow) showing nuclear overlapping, irregular contours, high nuclear/cytoplasmic ratios and anisocaryosis are readily detectable. B, sample from “Biopsy___13” (see Table 2): the specimen contains large amounts of mucus and cellular debris with few or no intact cells; original magnification, 200.

Endoscopic ultrasound (EUS) showed a low echoic mass less than 20mm in diameter within the pancreas in patient 12 (see Table 4). Biopsy specimens obtained by EUS-FNAB revealed well-differentiated adenocarcinoma in this patient. However, from the cytology, it was difficult to diagnose malignancy, because of the presence of welldifferentiated cancer cells. H&E, ¥120
Mucinous tumour, low power view. Cellular aspirate demonstrating abundant background mucus. Clusters of cells are embedded within the mucus. This could be from a mucinous cystic tumour or from intraductal papillary mucinous tumour. (Papanicolaou x10).

My approach to pancreatic fine needle aspiration
Gladwyn Leiman
J Clin Pathol 2007;60:43–
Mucinous cystic neoplasm, oil immersion view. A sheet of mucin producing columnar epithelial cells seen at very high magnification, demonstrating pale intra-cytoplasmic secretion, which is mucin.

Figure 2: Aggregates of pancreatic ductal carcinoma. Irregular or

My approach to pancreatic fine needle aspiration
Gladwyn Leiman
Neuro-endocrine tumour. This tightly packed cell cluster demonstrates one FNA pattern of neuro-endocrine or islet cell tumour. The cells are crowded in three-dimensional acini, and are characterized by granular chromatin. (Papanicolaou 40)

My approach to pancreatic fine needle aspiration
Gladwyn Leiman
Solid-cystic pseudopapillary neoplasm, fixed. On the fixed slide, the stromal tissue forming the core centres is virtually inapparent, giving a false impression of acinar formations. This may mimic islet cell tumour, but displays bland chromatin. (Papanicolaoux40).

My approach to pancreatic fine needle aspiration
Gladwyn Leiman
My approach to pancreatic fine needle aspiration
Gladwyn Leiman

Very rare pancreatic tumours
“Too few cases of serous cystadenoma have been described to enable confident cytological assessment.77 78 The only case I have encountered in 25 years was missed cytologically three times!”
The unknowns 1-5

• Look at the slides, and make your opinion
• Write it down for yourself
• At the end of this session I will show the outcome of the patients
“right diagnoses”

1. Severe dysplasia in explant
2. No follow up so far: A is answered inflammatory benign and B marked atypia, suspicious for dysplasia
3. Infection- Autopsy revealed TBC (Pathologist and Surgeon are now on medication because of x-ray findings)
4. Peripheral T-cell lymphoma in pancreas
5. Ductal cancer in corpus pancreatis