Microscopy of CSF & Body Fluids

Tracy I. George, MD
Professor of Pathology
University of Utah Health
Executive Director, Clinical Trials & PharmaDx
ARUP Laboratories

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Objectives

- Distinguish benign from malignancy cytology on CSF and body fluids
- Recommend appropriate further evaluation when necessary
Agenda

Item 1 Cytopreparatory Methods

Item 2 Cerebrospinal Fluid

Item 3 Pleural Fluid

Item 4 Peritoneal Fluid

Item 5 Pericardial fluid
Cytopreparatory Methods

- Centrifugation w/ preparation from sediment
- Cytocentrifugation
- ThinPrep™
- Membrane filtration
- Cell block preparation
  - Immunohistochemistry, in situ hybridization, molecular studies
Agenda

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Tubes

- Tube 1: Chemistry & immunologic studies
- Tube 2: Microbiologic studies
- Tube 3: Cell count & cytologic examination

✓ *In a traumatic tap, after centrifugation the supernatant fluid will clear*

✓ *+/- clot, no xanthochromia*
## Cerebrospinal Fluid

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Adult</th>
<th>Neonate</th>
</tr>
</thead>
<tbody>
<tr>
<td>WBC</td>
<td>&lt;5/μL</td>
<td>&lt;30/μL</td>
</tr>
<tr>
<td>RBC</td>
<td>Few</td>
<td>Variable</td>
</tr>
<tr>
<td>Lymphs</td>
<td>40-80%</td>
<td>5-35%</td>
</tr>
<tr>
<td>Monos</td>
<td>15-45%</td>
<td>50-90%</td>
</tr>
<tr>
<td>PMNs</td>
<td>0-6%</td>
<td>0-8%</td>
</tr>
</tbody>
</table>

*Correction for bloody tap is usually 1-2 wbc/1,000 rbc*
Lymphocytes
Cns lymp

Monocytes
Benign vs. Malignant

**N:C ratio**
- **Low to moderate**
- **High**

**Nuclear contour/nuclear membrane**
- *Round to oval nucleus with a regular nuclear contour; prominent and distinct nuclear membrane*
- Irregular nuclear shape; indistinct nuclear membrane, may have “blebs” at periphery
Benign or Malignant?
Choroid Plexus Cells
Benign vs. Malignant

Nuclear texture

• Fine or uncondensed nuclear chromatin with uniform, smooth texture

• Uneven nuclear texture; prominent parachromatin spaces may occur

Nucleoli

• Small to medium-sized, if any are present

• Large and angulated nucleoli
Diagnosis?
Ependymal Cells
Benign or malignant?
CNS ganglion cells
Diagnosis?
Bone marrow contamination
Bacterial Meningitis
Diagnosis?
Cryptococcus neoformans
Benign vs. Malignant

**Lymphoid cells**

- Heterogeneous with different morphologic forms
- Round to bean-shaped nuclei with regular contours
- Clear Golgi zone often present

- Homogeneous infiltrate of malignant cells
- Nuclei may have uneven contours
- Golgi region often absent in lymphoma cells
Diagnosis?
Viral meningitis
Benign vs. Malignant

**Lymphoid cells**

- **Nuclear chromatin varies from condensed to blastic** *(in the immunoblast)*
- **Several small nucleoli may be present**

- **Blastic nuclear chromatin in all cells**
- **Nucleoli may be large**
Diagnosis?
Acute Leukemia
Benign vs. Malignant

**Cell clusters**

- **Benign cells have thin spaces ("windows")**
- **Outer border of cell cluster is discontinuous**

- 3-D clusters with morula-like appearance
- Outer border of cell cluster is continuous and smooth, and may be darkly stained due to cell overlap
Benign or malignant?
Medulloblastoma
Large Cell Lymphoma

TP-Pap

CS-WG

1000x oil
Acute Lymphoblastic Leukemia

TP-Pap

CS-WG

1000x oil
Agenda

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Item 4 Pleural Fluid

Item 5 Peritoneal Fluid
### Transudates vs. Exudates

<table>
<thead>
<tr>
<th></th>
<th>≤ 0.5</th>
<th>&gt;0.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluid protein/serum protein</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fluid LDH/serum LDH</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LDH</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- **Clear**
- **Cloudy**
- **Pale yellow**
- **Turbid**
- **Purulent**
- ** Bloody**

- **Benign**
- **Malignant**
- **Infection**
Pericardial Fluid

• Constitute ~ 1% serous effusions

• *Most are exudates*
  – malignancy; infection; trauma; MI; leaking aneurysm; RA; SLE; anti-coag therapy

• Transudates
  Congestive heart failure; RA; sarcoidosis; hypothyroidism

• Lymphocytosis:
  Adult: **TB**
  Child: **Viral**
  ✓ Rarely lymphoma
Sources of Malignancy in Pericardial Fluid

**Malignant cytology:** 27%

1. Breast: 33%
2. Colon: 20%
3. Lung: 20%
4. Lymphoma/leukemia: 7%
5. Misc/Unknown: 20%

Zipf RE. *The role of cytology in the evaluation of pericardial fluids.* Chest 1972;62:593-596
Etiology of Pleural Effusions

- Malignancy: 25% - 45%
- CHF: 12%
- Infection: 22% (TB, Bacterial, Viral, Fungal, etc)
- Indeterminant: 10%
- Pulmonary embolism/Infarct: 3%
- Cirrhosis: 2%
- Collagen Dz: 2%

Hausheer F. *Dx & Treatment of malignant pleural effusions*. Semin Oncol. 1985;12(1):54-75
Pleural effusions

Transudate

- CHF
- Cirrhosis
- Hypoproteinemia
- Nephrotic syndrome
- Atelectasis
- Myxedema
- Peritoneal dialysis
- PE
- Meig’s syndrome
- Obstructive uropathy

Exudate

- Malignancy
- Infection
- Trauma
- Pulmonary infarction
- PE
- Autoimmune dz
- Pancreatitis
- Ruptured esophagus
# Peritoneal Effusions

**Transudate**
- CHF
- Cirrhosis
- Hypoproteinemia
- Nephrotic syndrome
- Hepatic vein occlusion
- Hepatic metastasis

**Exudate**
- Malignancy
- Infection
- Trauma
- Pancreatitis
- Bile peritonitis
# Malignant Ascitic Fluid

## Male

<table>
<thead>
<tr>
<th>Category</th>
<th>Percentage Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Misc</td>
<td>38 - 46.4%</td>
</tr>
<tr>
<td>GI tract</td>
<td>23.8 - 42.8%</td>
</tr>
<tr>
<td>Unknown</td>
<td>14.2 - 16.6%</td>
</tr>
<tr>
<td>Lung</td>
<td>4.7 - 9.5%</td>
</tr>
</tbody>
</table>

## Female

<table>
<thead>
<tr>
<th>Category</th>
<th>Percentage Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genital Tract</td>
<td>50.6 - 61%</td>
</tr>
<tr>
<td>Misc.</td>
<td>13.2 - 15.5%</td>
</tr>
<tr>
<td>Breast</td>
<td>10.3 - 17.6%</td>
</tr>
<tr>
<td>GI Tract</td>
<td>9.0 - 9.7%</td>
</tr>
<tr>
<td>Unknown</td>
<td>3.8 - 8.3%</td>
</tr>
<tr>
<td>Lung</td>
<td>0 - 1.7%</td>
</tr>
</tbody>
</table>
Macrophage & Mesothelial Cells
Macrophages
Multinucleated Giant Cells
Pigment Laden Macrophages
Mesothelium
Mesothelial Cells
Mesothelial Cells
Mesothelial Variability
Signet Ring Mesothelials
Mesothelial Windows
Mesothelial Hyperplasia
“Embracing” Mesothelials
Mesothelial Clumps
Mesothelial Mitoses
Mesothelial erythrophagocytosis
Diagnosis?
Malignant Mesothelioma
Immunohistochemistry

**Mesothelioma**
- Calretinin
- CK 5/6
- Two toned cytoplasm

**Adenocarcinoma**
- CEA
- CD15
- Ber EP-4
- B72.3
- Homogeneously distributed stain
Calretinin immunostain
Electron Microscopy

**Mesothelioma**

**Adenocarcinoma**
Benign vs. Malignant

Unusual homogenous population

- Unusual cells may be present, but there is a heterogeneity of morphologic features, with a gradation of normal to reactive cells

- Atypical, distinct population of bizarre cells are seen that do not resemble any known benign variant
Lymphocytes
Diagnosis?
Large cell lymphoma
Diagnosis?
Burkitt lymphoma
Diagnosis?
Primary Effusion Lymphoma
## Features of Adenocarcinoma

<table>
<thead>
<tr>
<th><strong>Cell groups</strong></th>
<th><strong>Individual cells</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>✓ Solid cell balls</td>
<td>✓ Signet-ring forms</td>
</tr>
<tr>
<td>✓ Papillary forms</td>
<td>✓ Intracytoplasmic lumena</td>
</tr>
<tr>
<td>✓ Free-floating acini</td>
<td>✓ Clear cell change</td>
</tr>
<tr>
<td>✓ Columnar cell fronds</td>
<td>✓ Intracytoplasmic dot</td>
</tr>
<tr>
<td>✓ Single-cell rows</td>
<td>✓ Multinucleated giant cells</td>
</tr>
</tbody>
</table>
Breast carcinoma
Diagnosis?
Breast Carcinoma
Ovarian Carcinoma
Papillary Serous Carcinoma
Diagnosis?
Lung Adenocarcinoma
Diagnosis?
Breast Carcinoma
Benign vs. Malignant

**Cytoplasmic vacuoles**

- *Tiny, poorly defined vacuoles can be seen in any degenerating cell*
- *Phagocytic vacuoles*
- *Mesothelial cells*

- Vacuoles in malignant cells usually represent synthesized material (e.g. mucin)
  - Adenocarcinoma often has large vacuoles with a smooth, well-defined border and a clear interior. Vacuoles may coalesce in adjacent cells
Pancreatic Adenocarcinoma
Breast Carcinoma
Gastric Adenocarcinoma
Conclusions

• **Benign vs. Malignant Criteria**
  • Nuclear Contour
  • Nuclear texture
  • Nucleoli
  • N:C ratio
  • Mitoses
  • Nuclear molding
  • Cytoplasmic vacuoles & granules
  • Signet-ring cells
  • Cell clusters
  • Unusual population
  • Reactive vs malignant lymphoid cells
Recommended Reading

- CAP Color Atlas of Body Fluids
- Kjeldsberg Body Fluids