

Microscopy of CSF & Body Fluids

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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

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Tubes

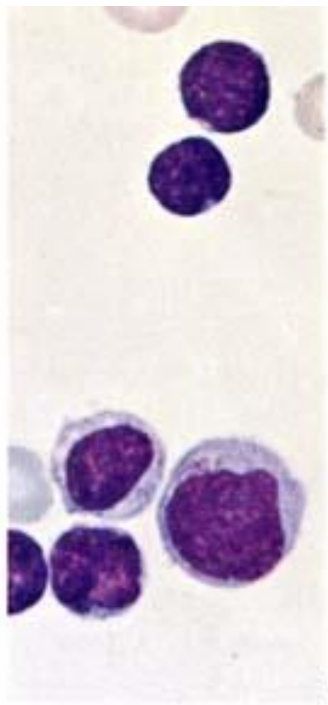
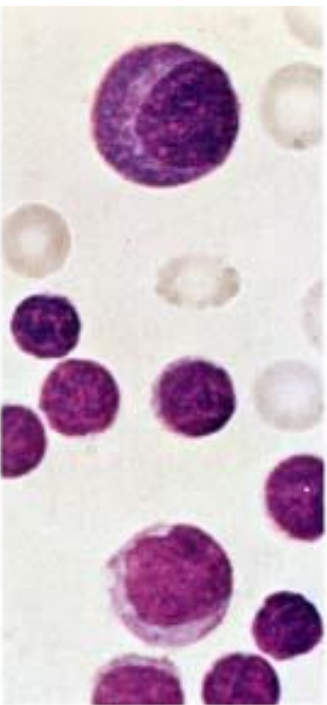
- **Tube 1: Chemistry & immunologic studies**
 - **Tube 2: Microbiologic studies**
 - **Tube 3: Cell count & cytologic examination**
- ✓ *In a traumatic tap, after centrifugation the supernatant fluid will clear*
- ✓ *+/- clot, no xanthochromia*

Cerebrospinal Fluid

<u>Cell type</u>	<u>Adult</u>	<u>Neonate</u>
WBC	<5/μL	<30/μL
RBC	Few	Variable
Lymphs	40-80%	5-35%
Monos	15-45%	50-90%
PMNs	0-6%	0-8%

Correction for bloody tap is usually 1-2 wbc/1,000 rbc

Lymphocytes
Cns lymph



Monocytes



Benign vs. Malignant

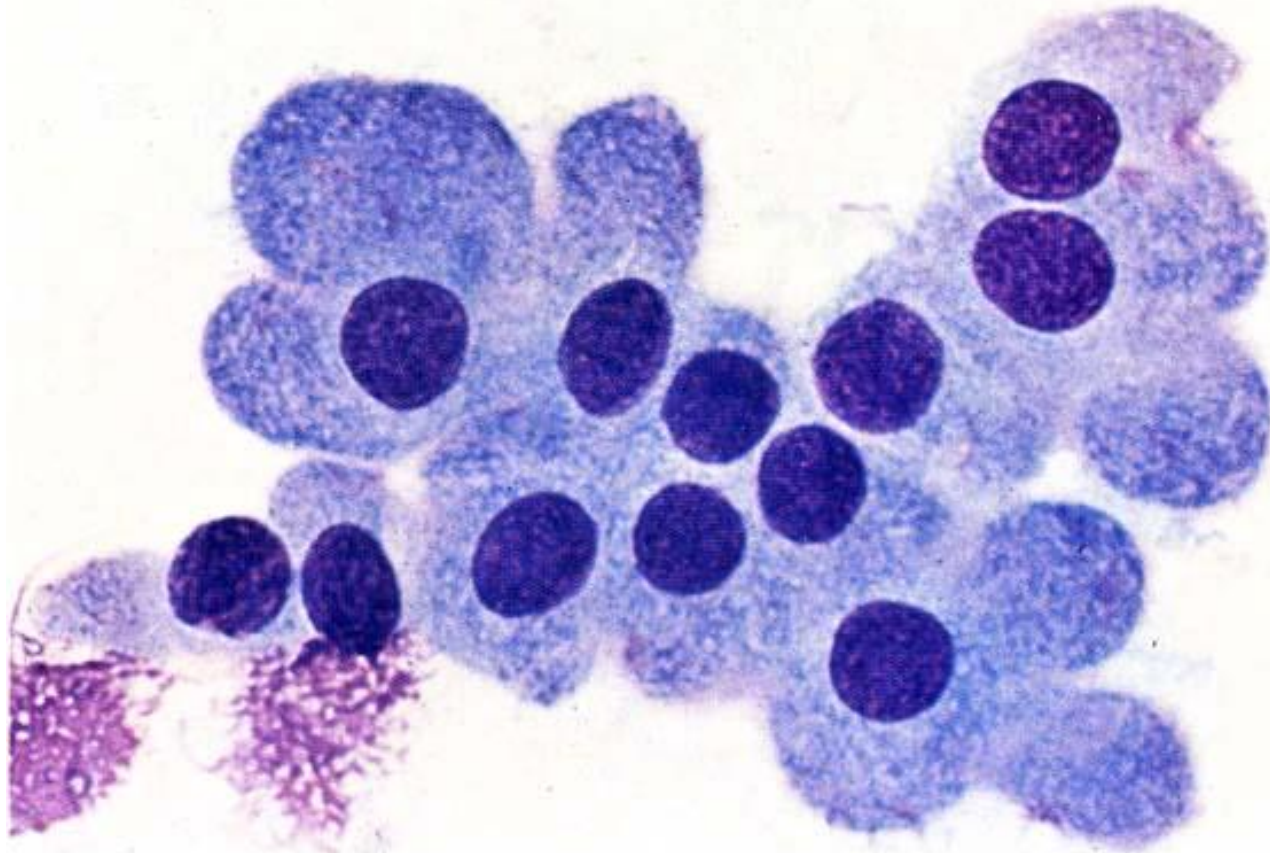
N:C ratio

- *Low to moderate*
- High

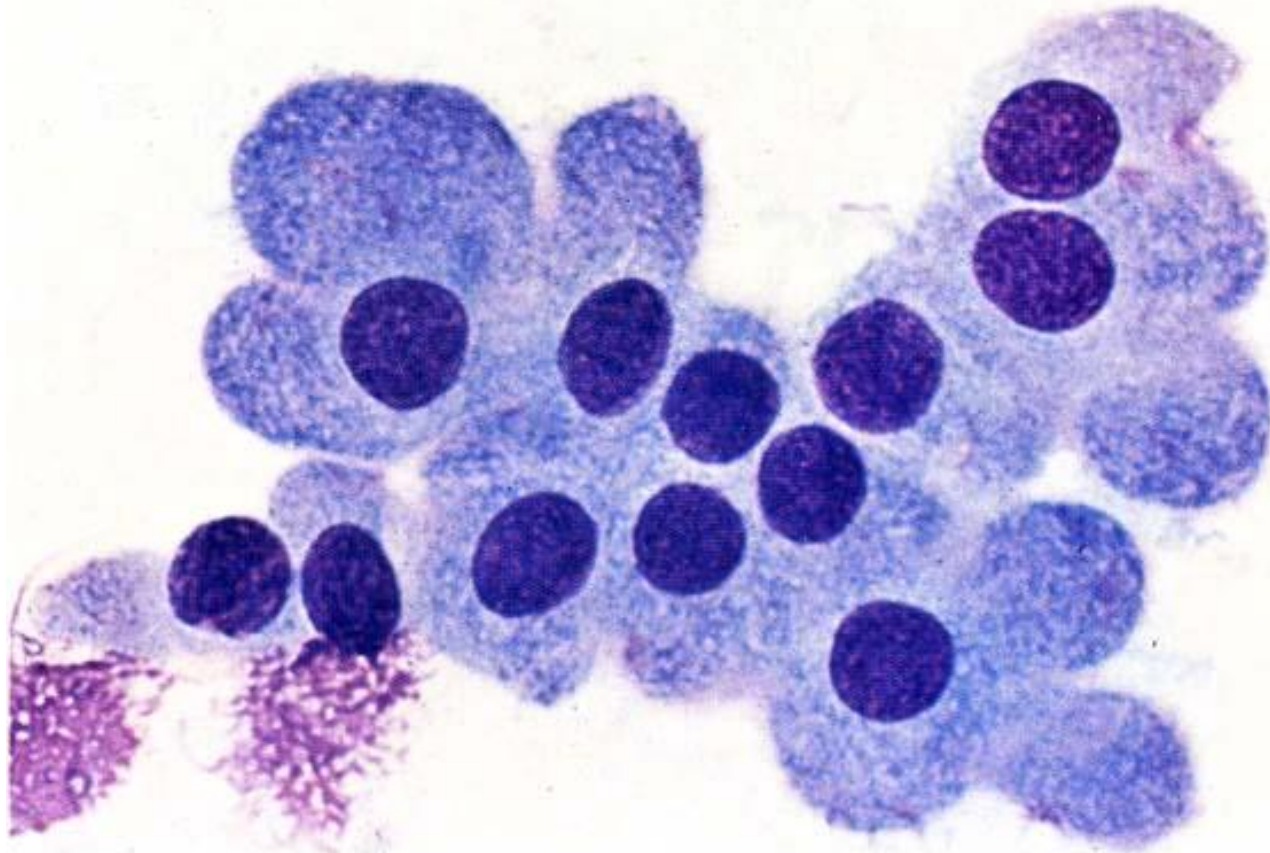
Nuclear contour/nuclear membrane

- *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

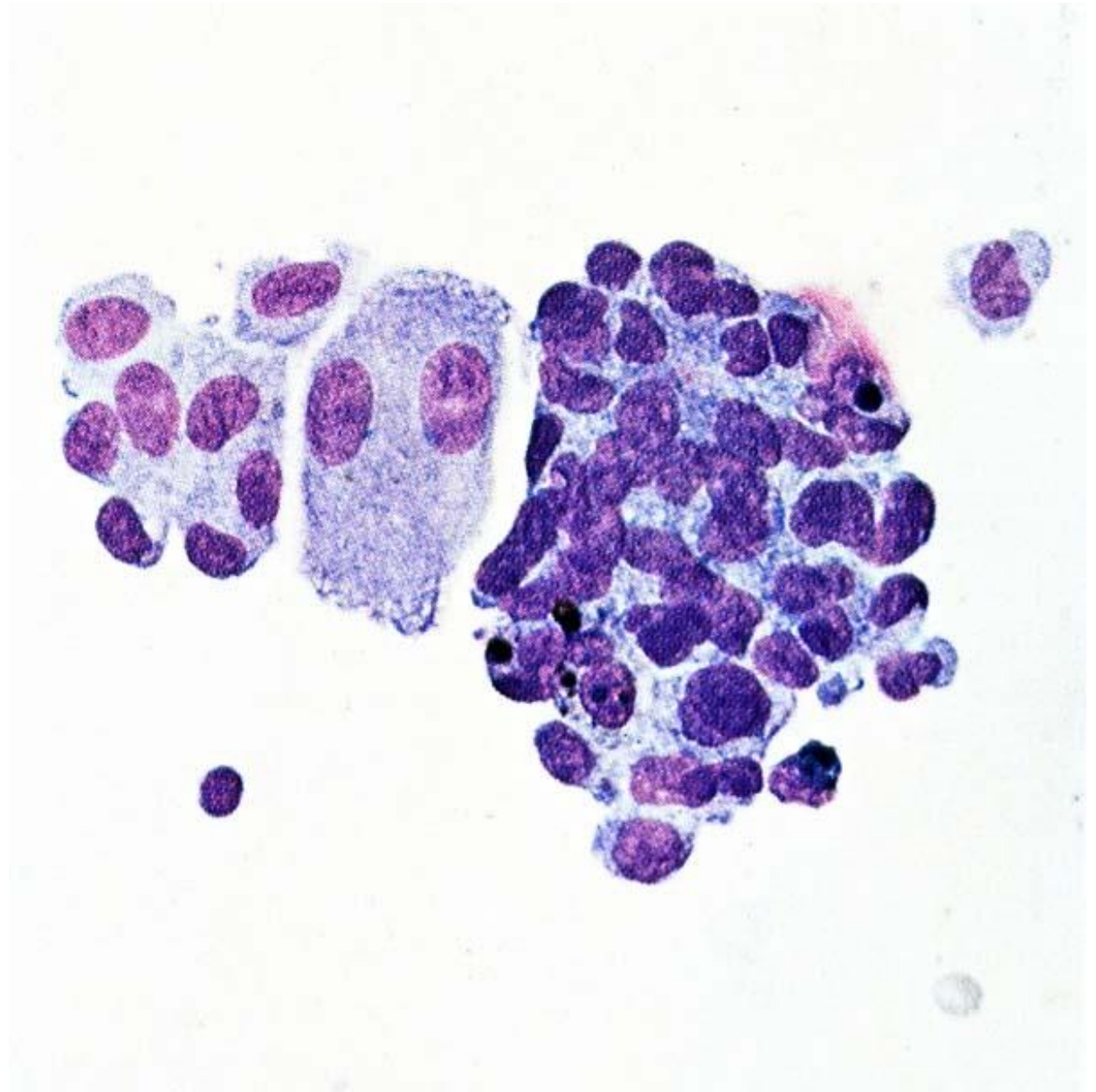
Nuclear texture

- ***Fine or uncondensed nuclear chromatin with uniform, smooth texture***
- Uneven nuclear texture; prominent parachromatin spaces may occur

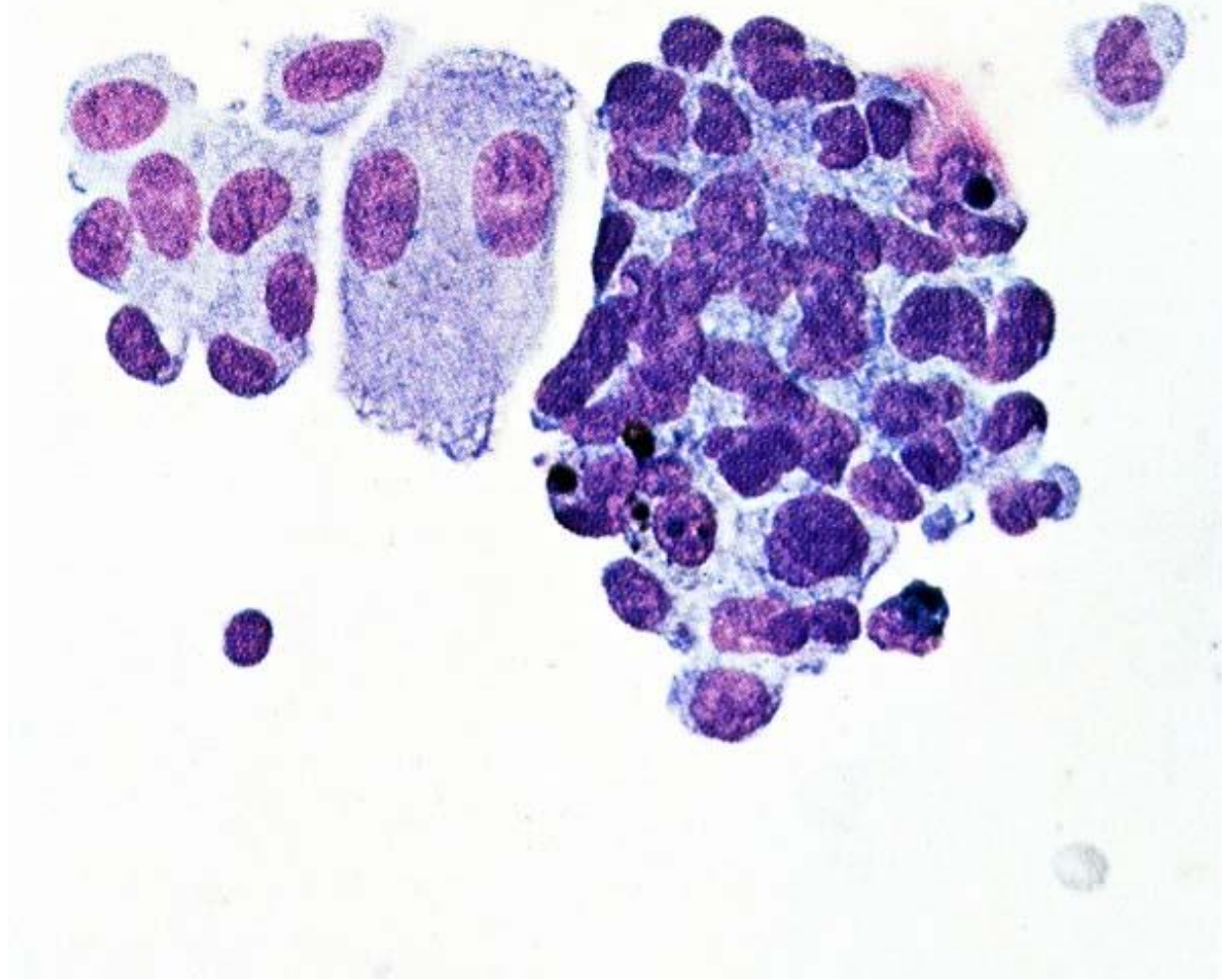
Nucleoli

- ***Small to medium-sized, if any are present***
- Large and angulated nucleoli

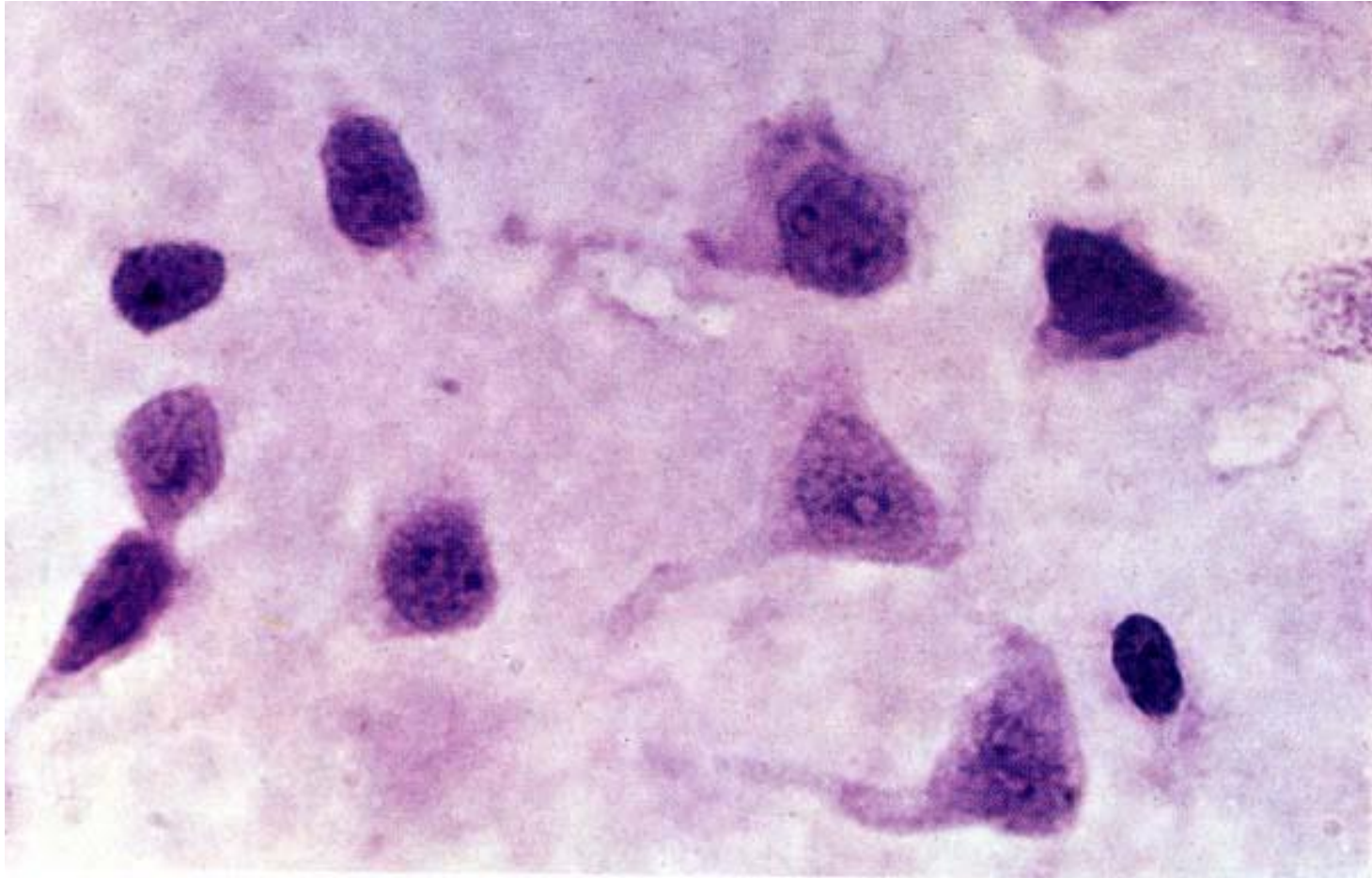
Diagnosis?



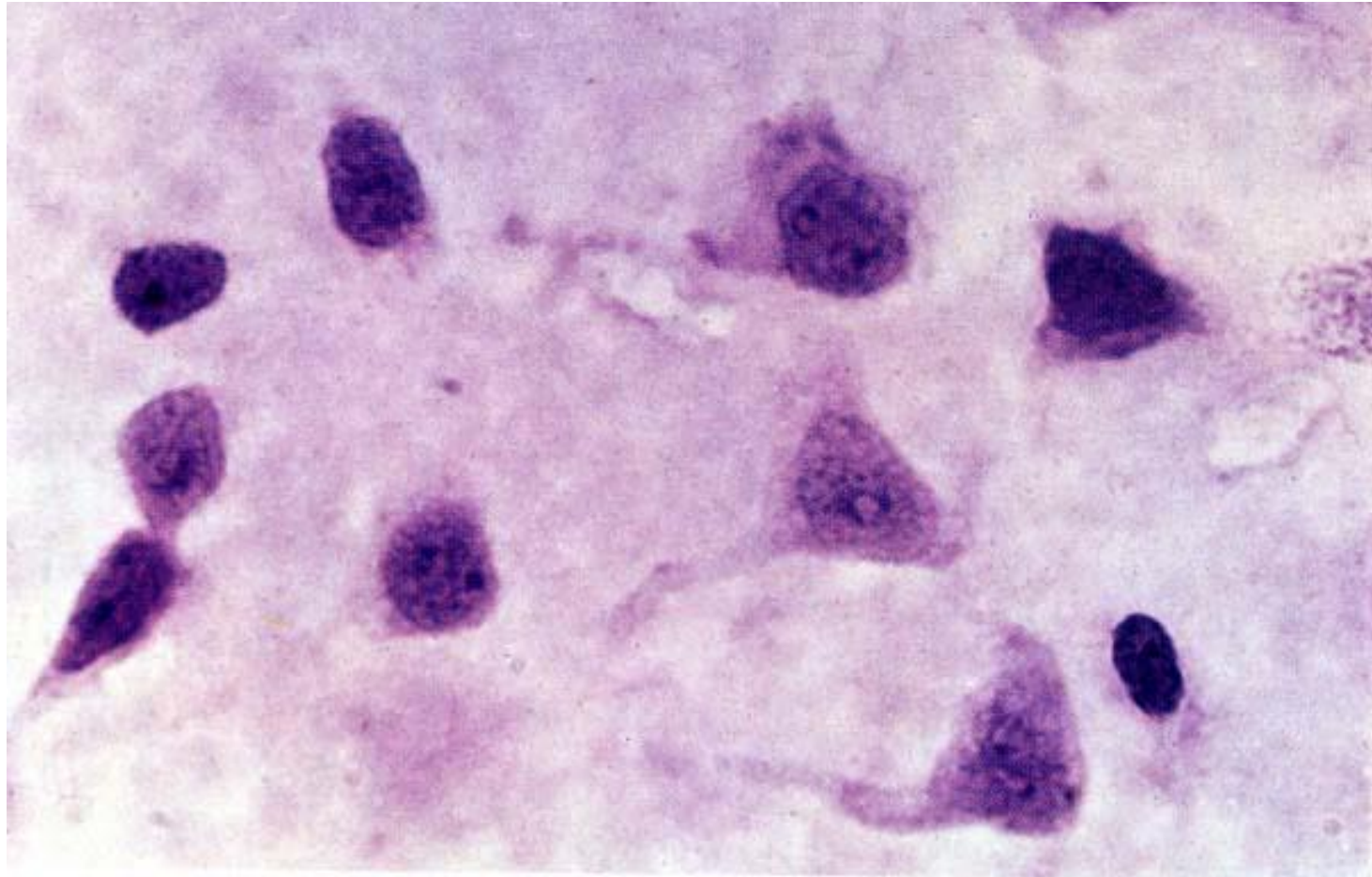
Ependymal Cells



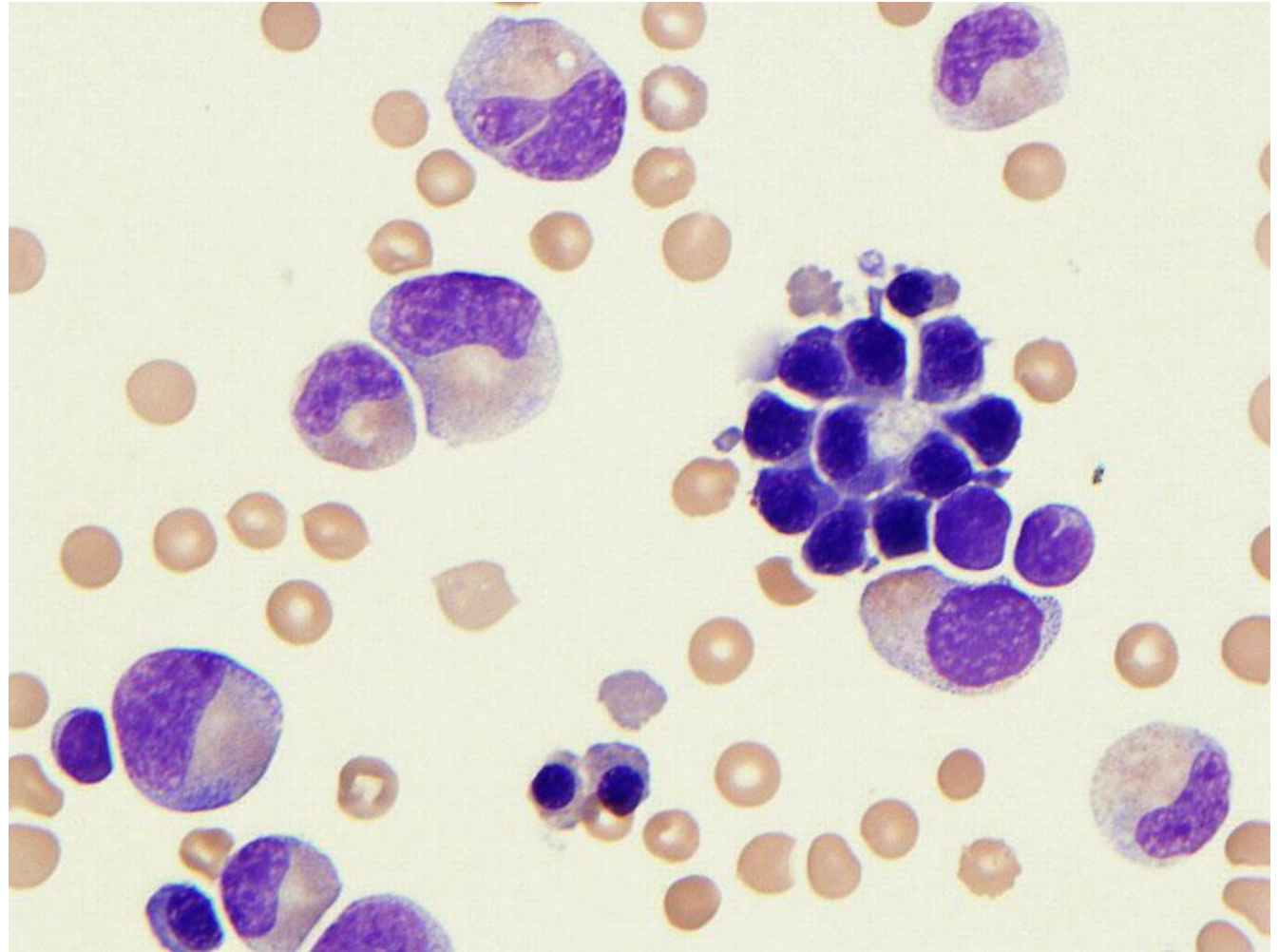
Benign or malignant?



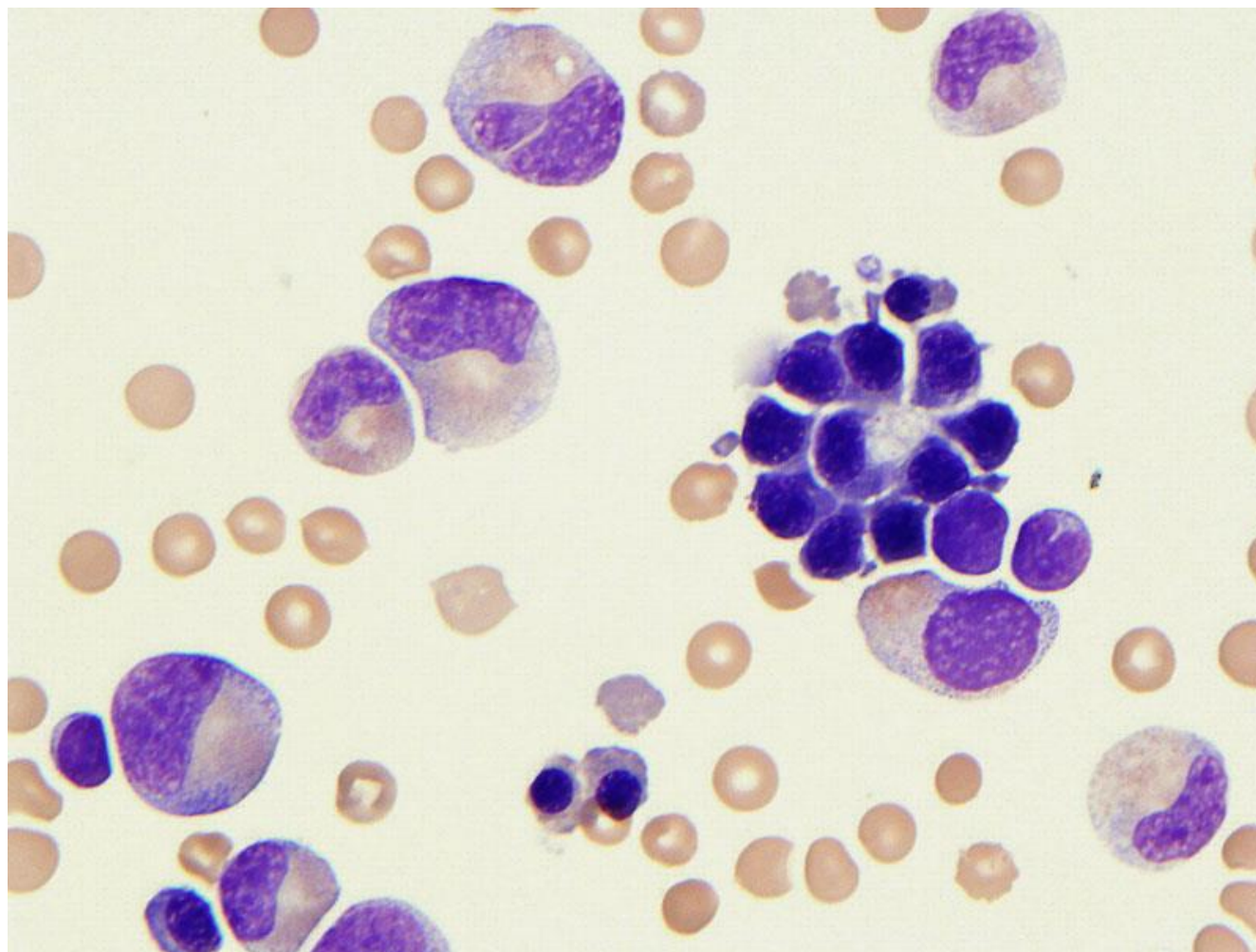
CNS ganglion cells



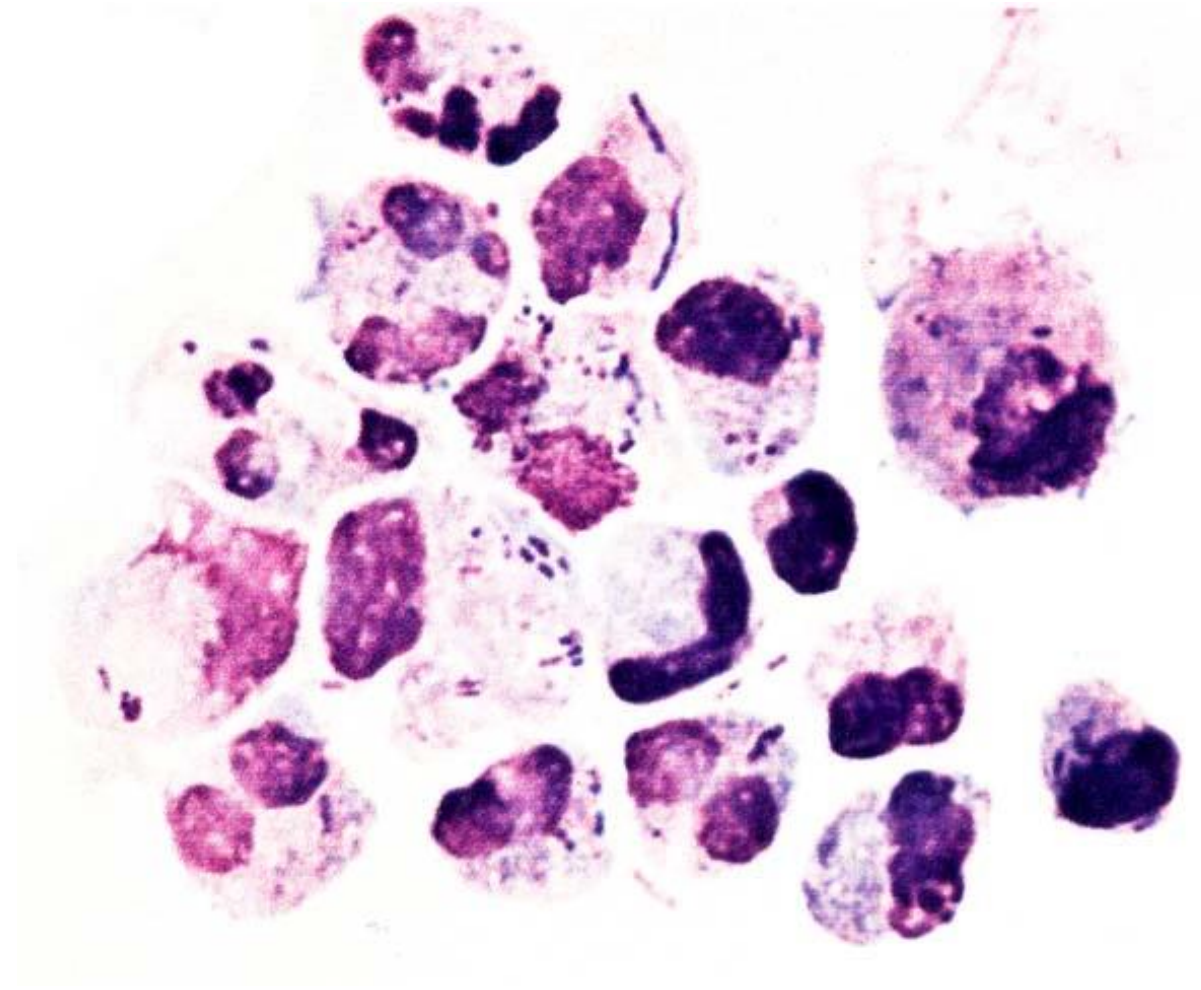
Diagnosis?



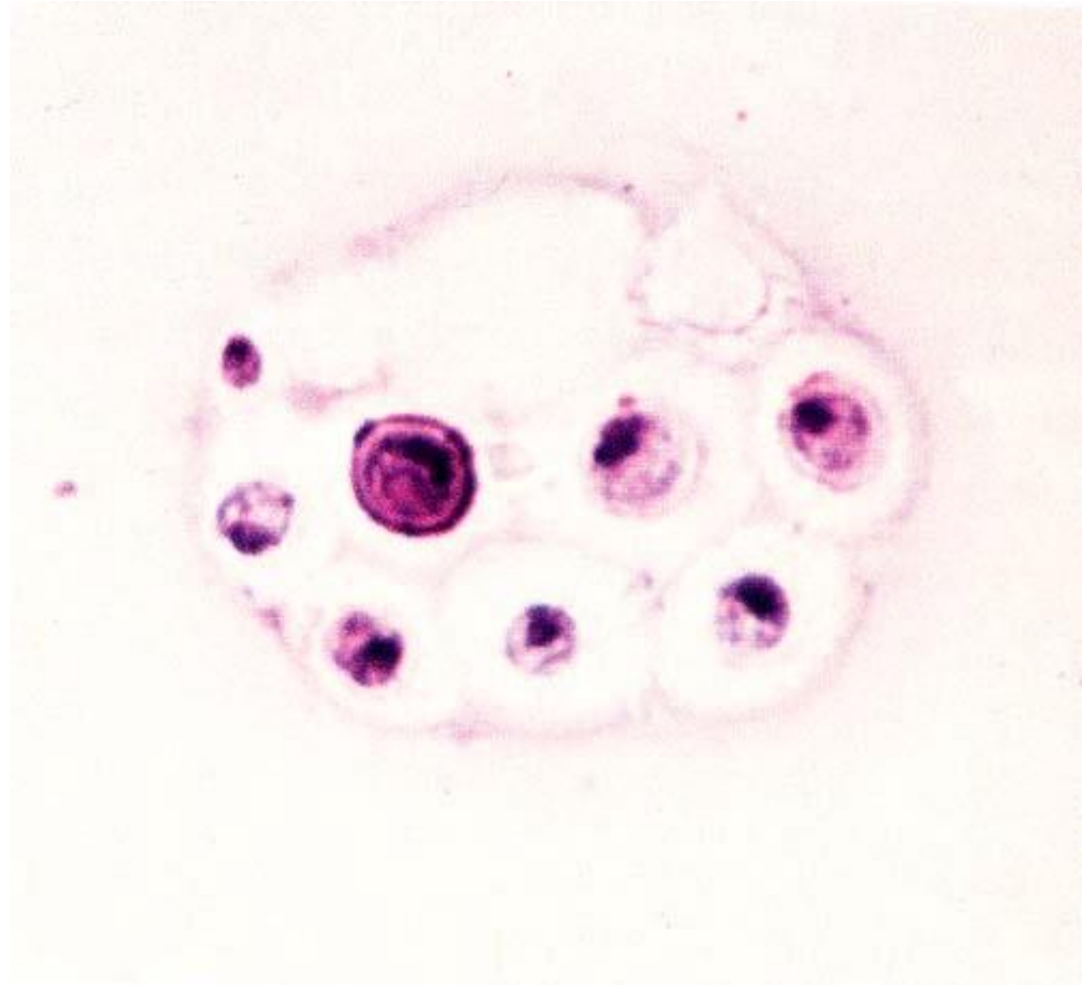
Bone marrow contamination



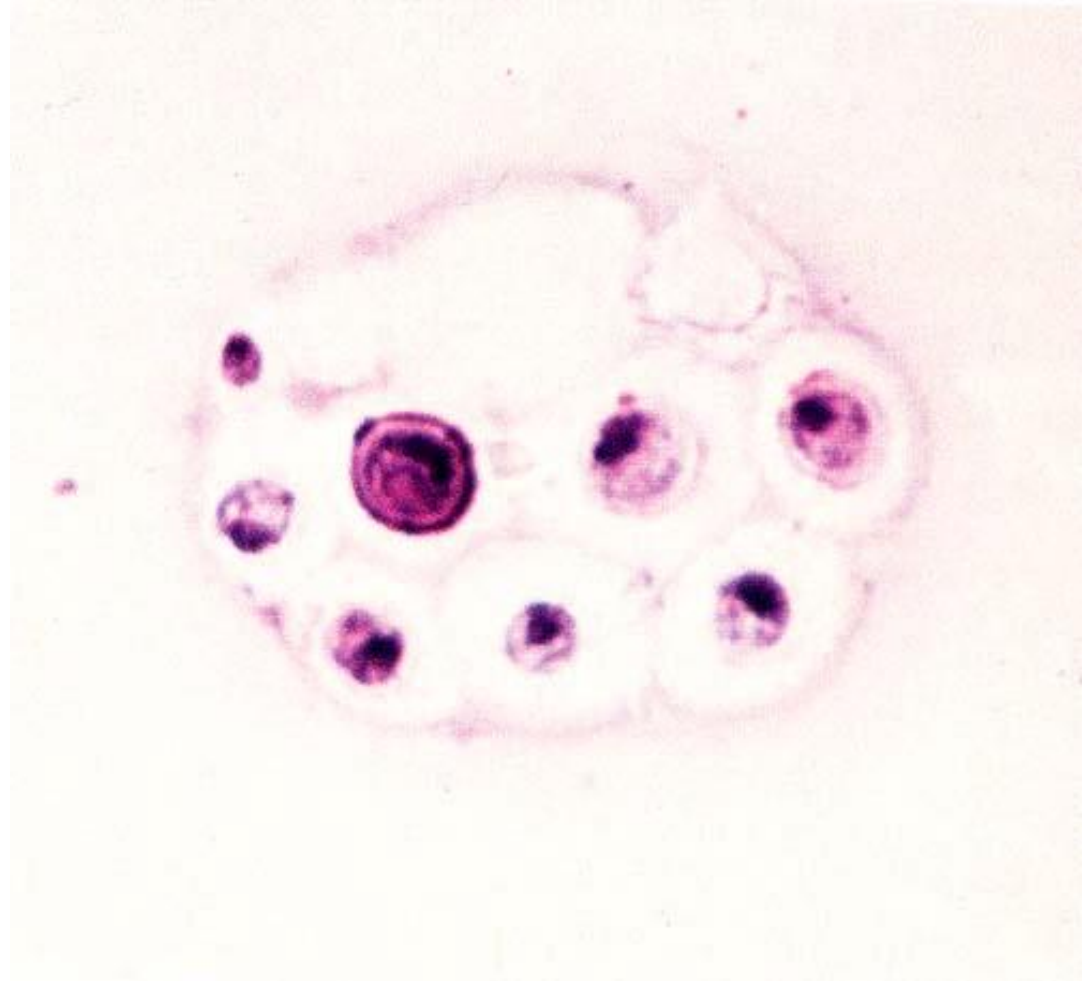
Bacterial Meningitis



Diagnosis?



Cryptococcus neoformans

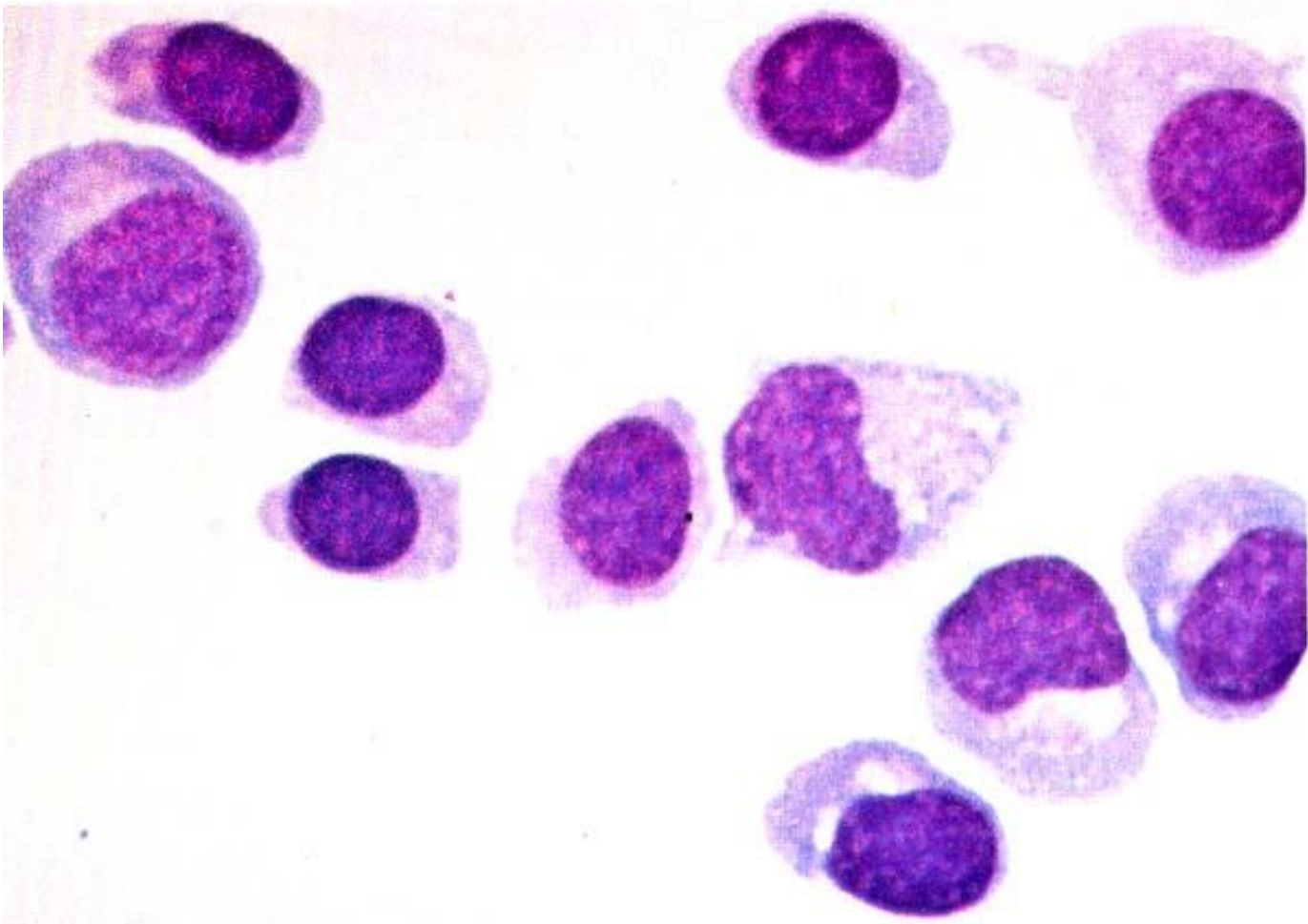


Benign vs. Malignant

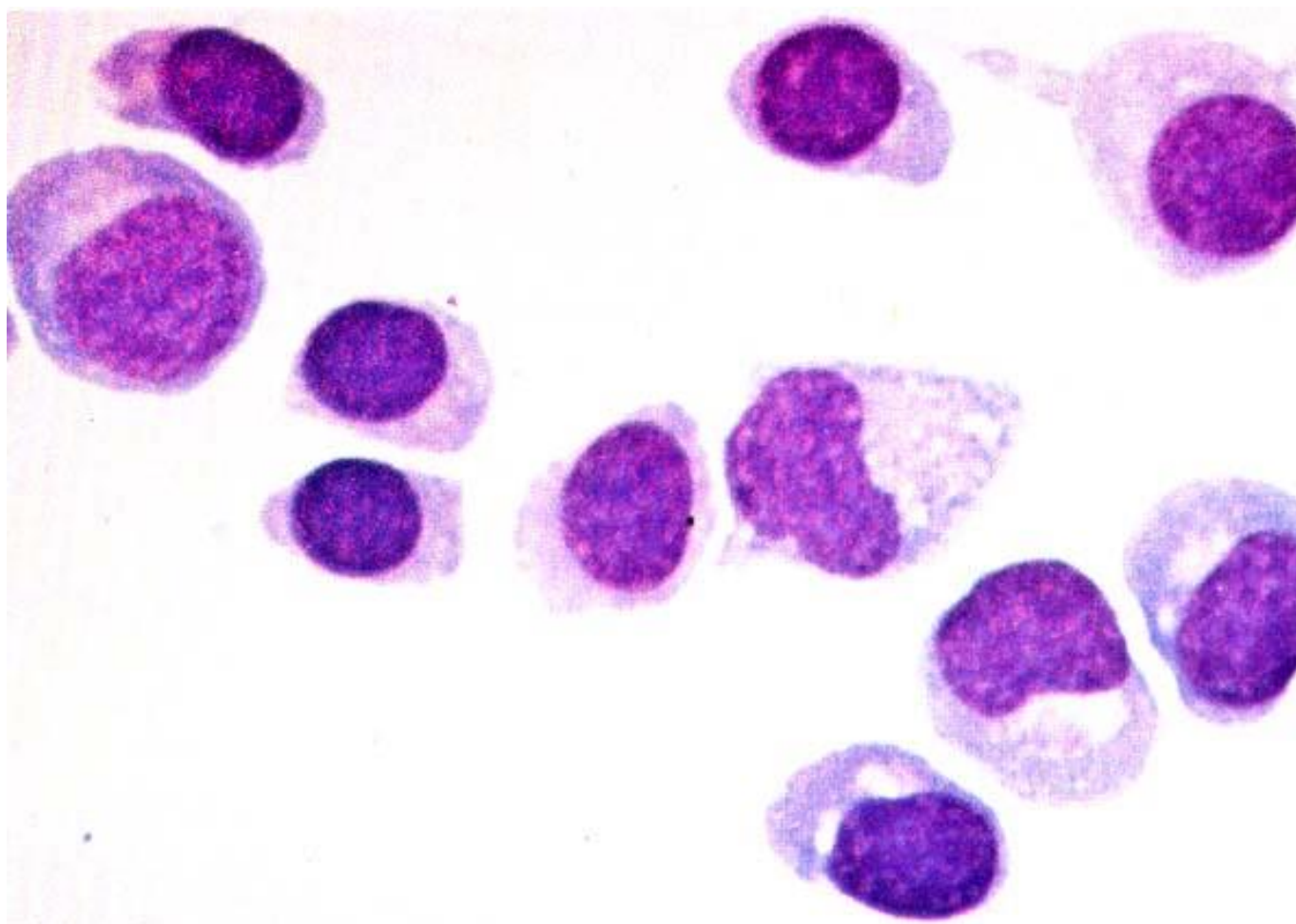
Lymphoid cells

- ***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

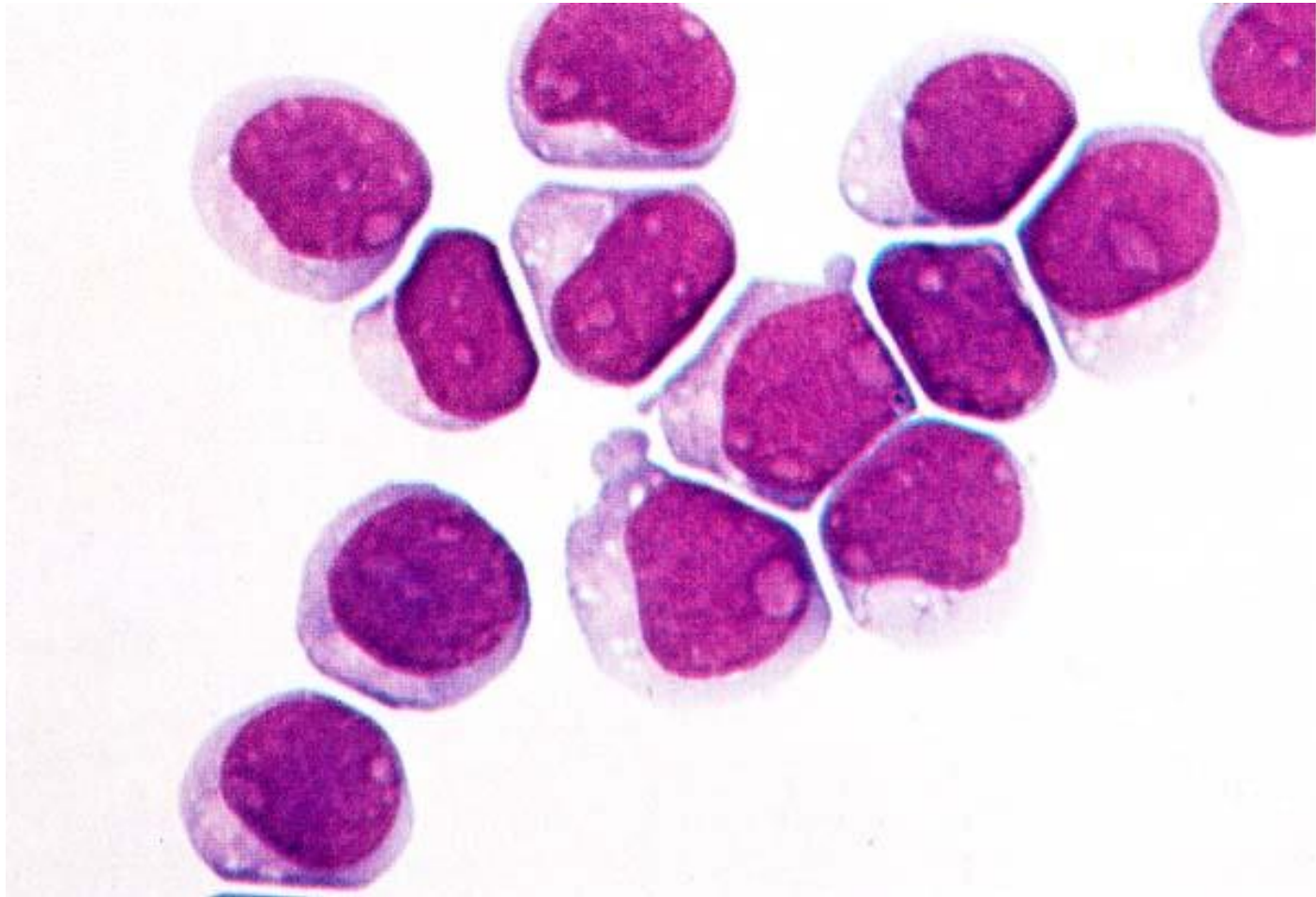


Benign vs. Malignant

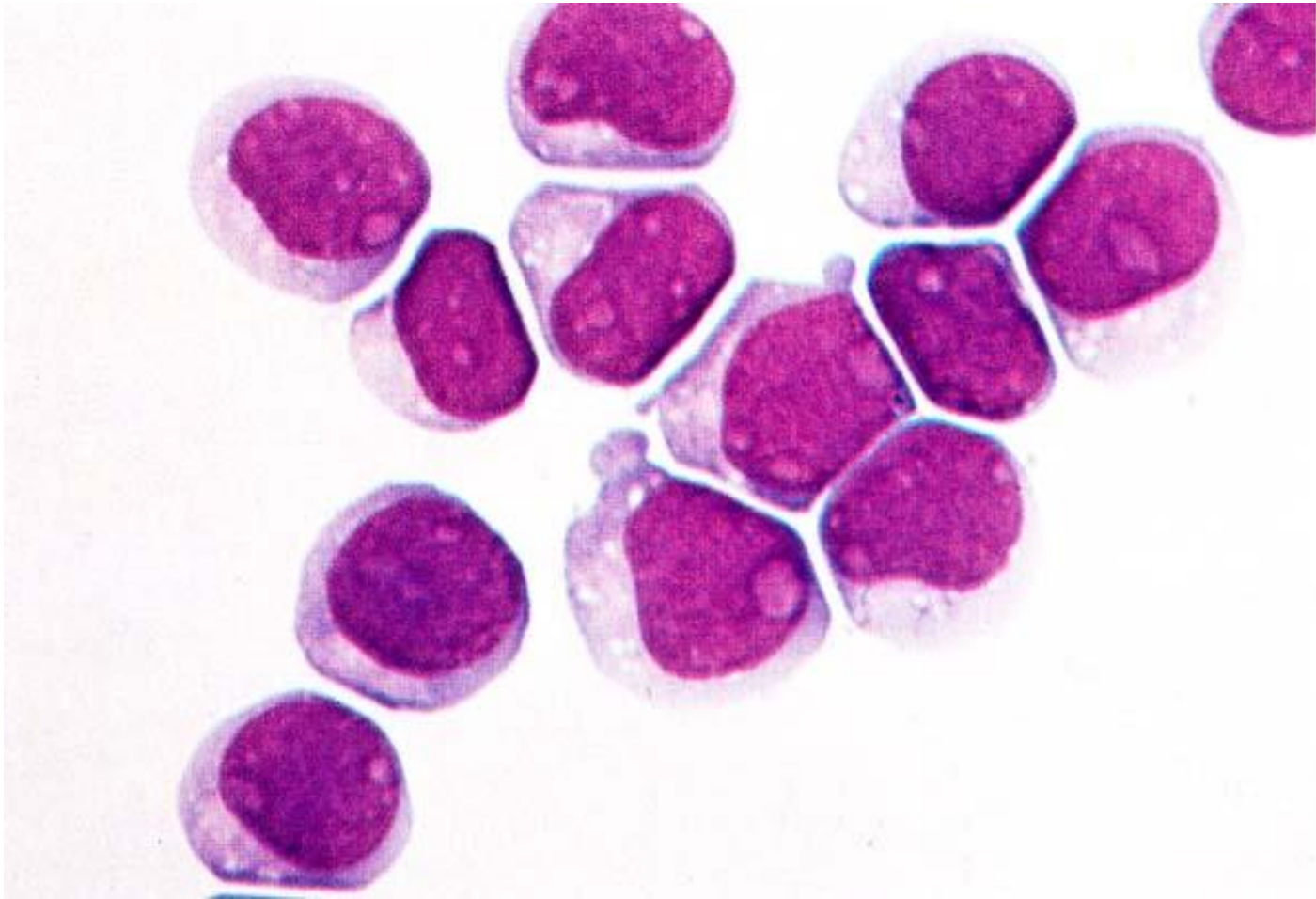
Lymphoid cells

- ***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

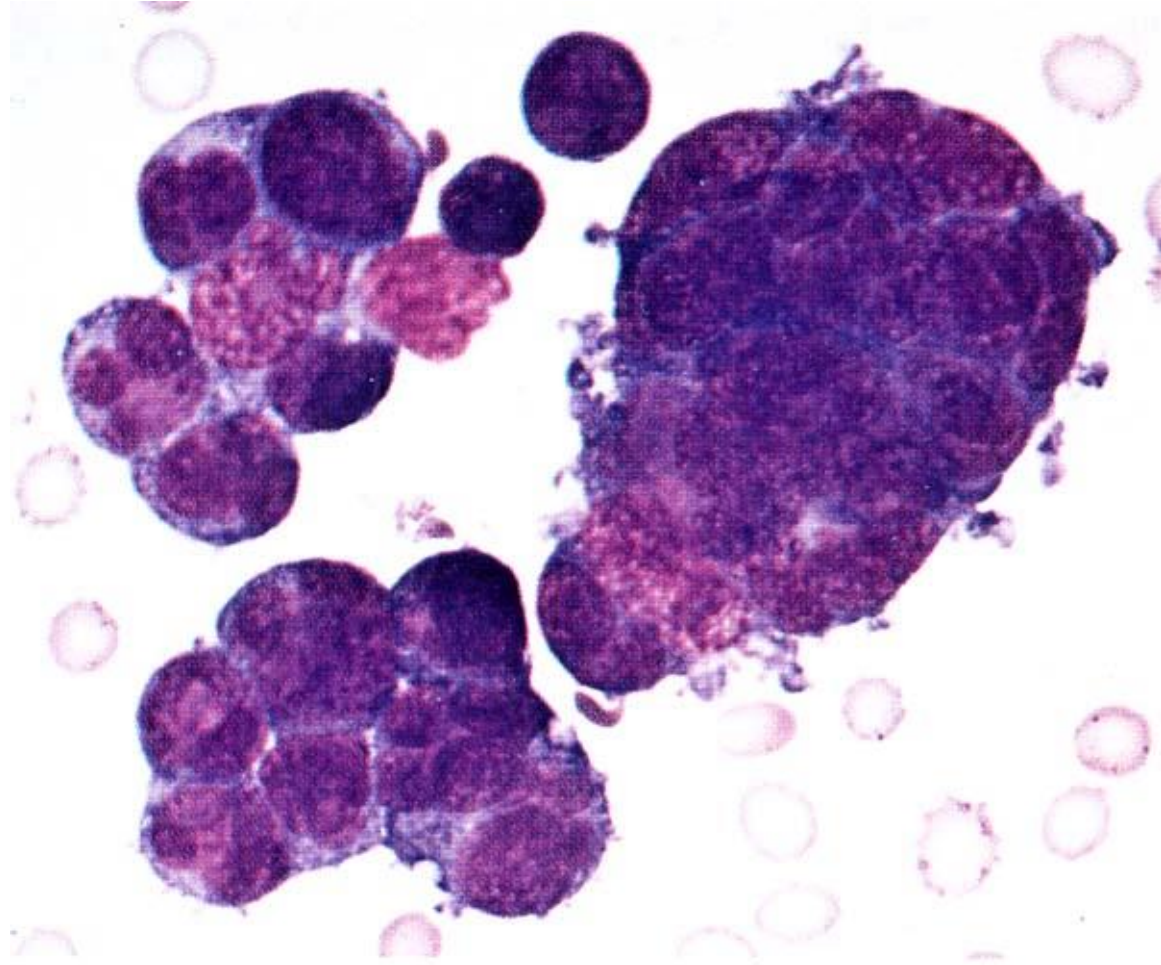


Benign vs. Malignant

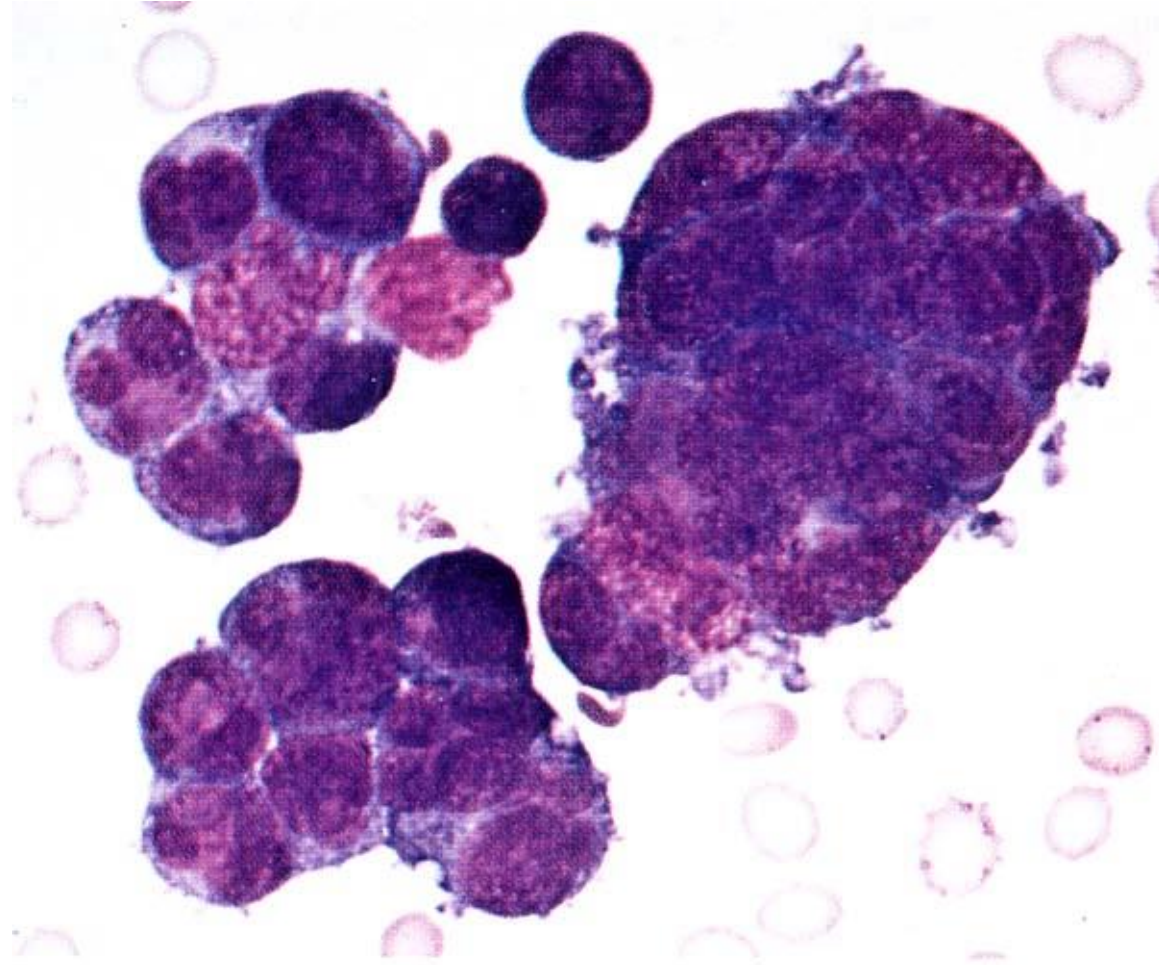
Cell clusters

- ***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?

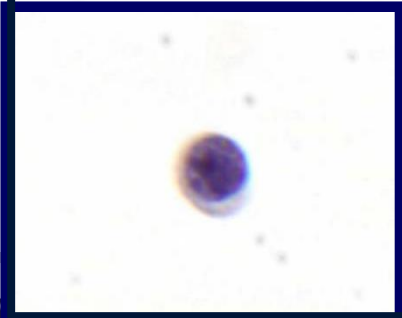
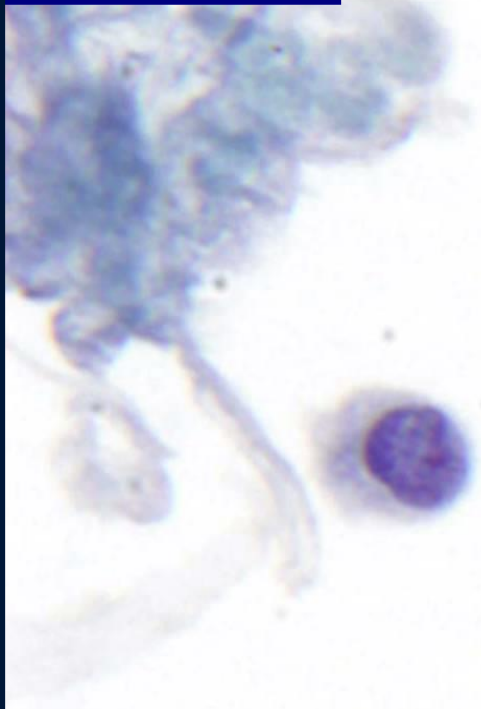


Medulloblastoma

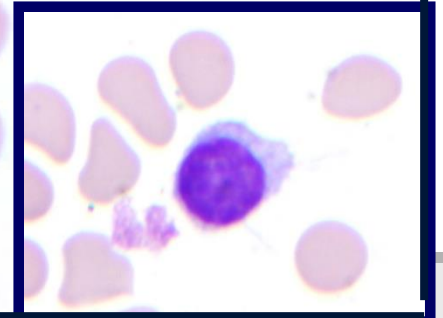


Large Cell Lymphoma

TP-Pap



CS-WG



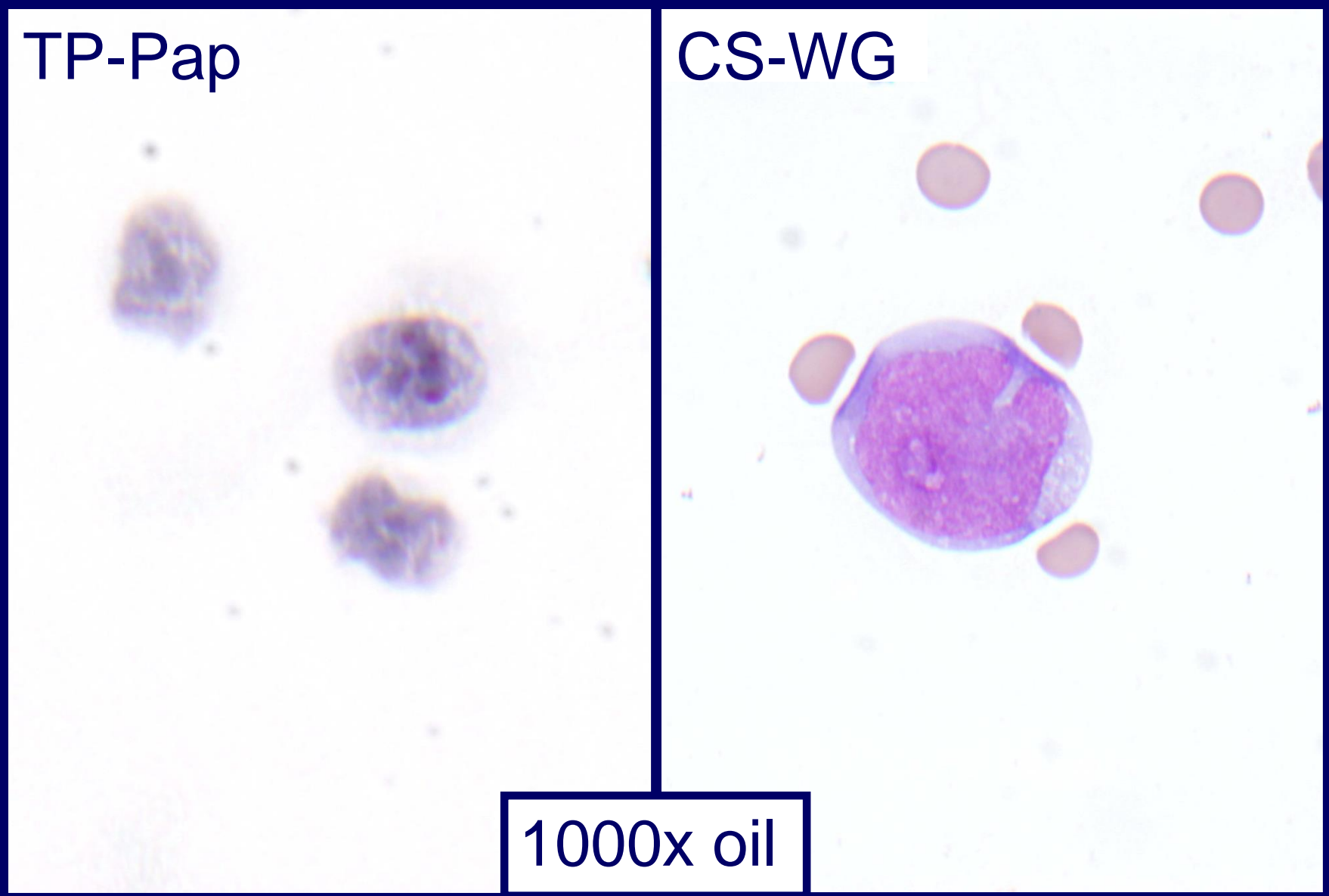
1000x oil

Acute Lymphoblastic Leukemia

TP-Pap

CS-WG

1000x oil



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Transudates vs. Exudates

• Fluid protein/serum protein	≤ 0.5	>0.5
• Fluid LDH/serum LDH	≤ 0.6	>0.6
• LDH	≤ 200	>200
	clear	cloudy
	pale yellow	turbid
		purulent
		bloody
	<i>benign</i>	<i>malignant</i>
		<i>infection</i>

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

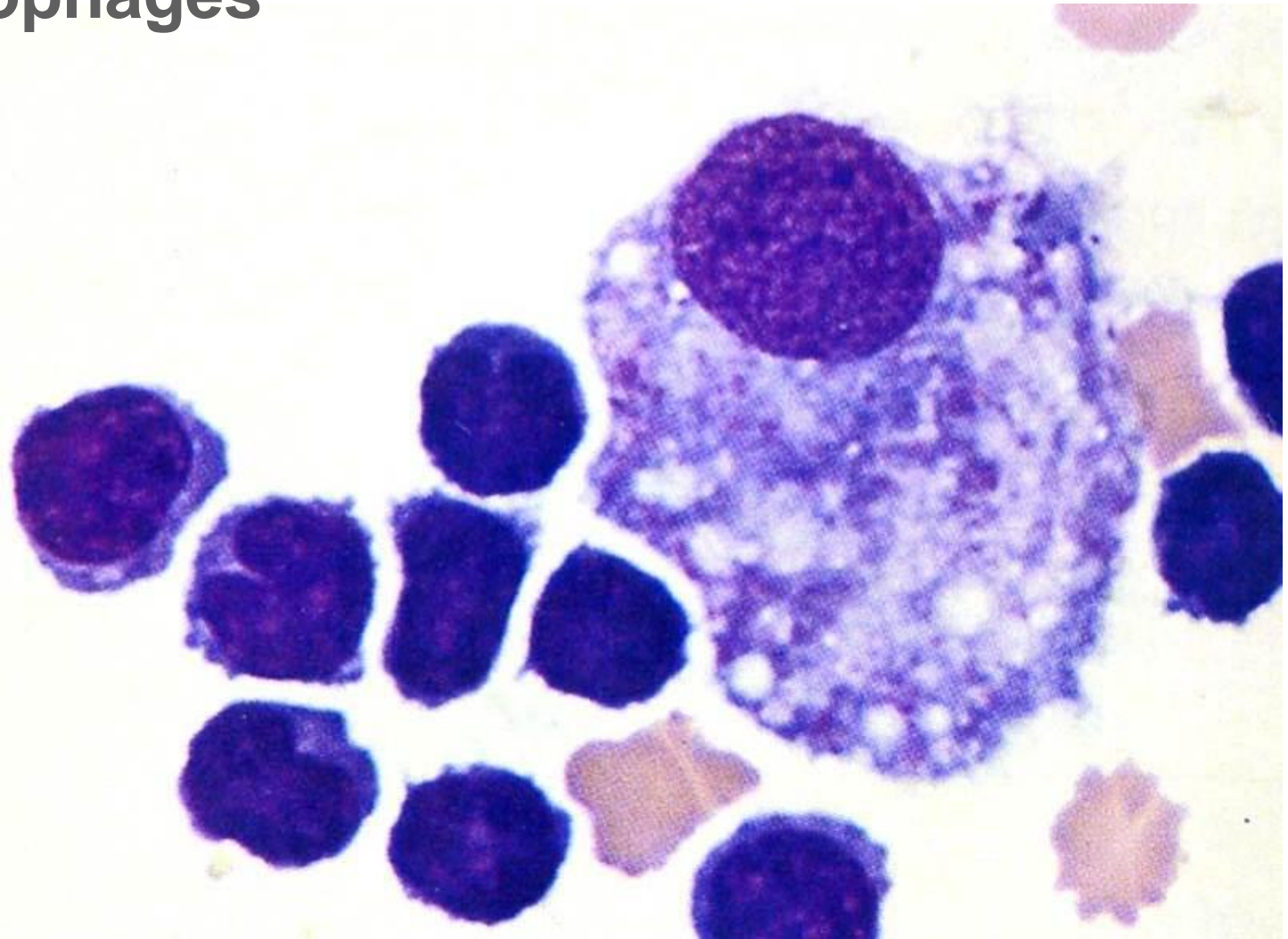
Misc	38 - 46.4%
GI tract	23.8 - 42.8%
Unknown	14.2 - 16.6%
Lung	4.7 - 9.5%

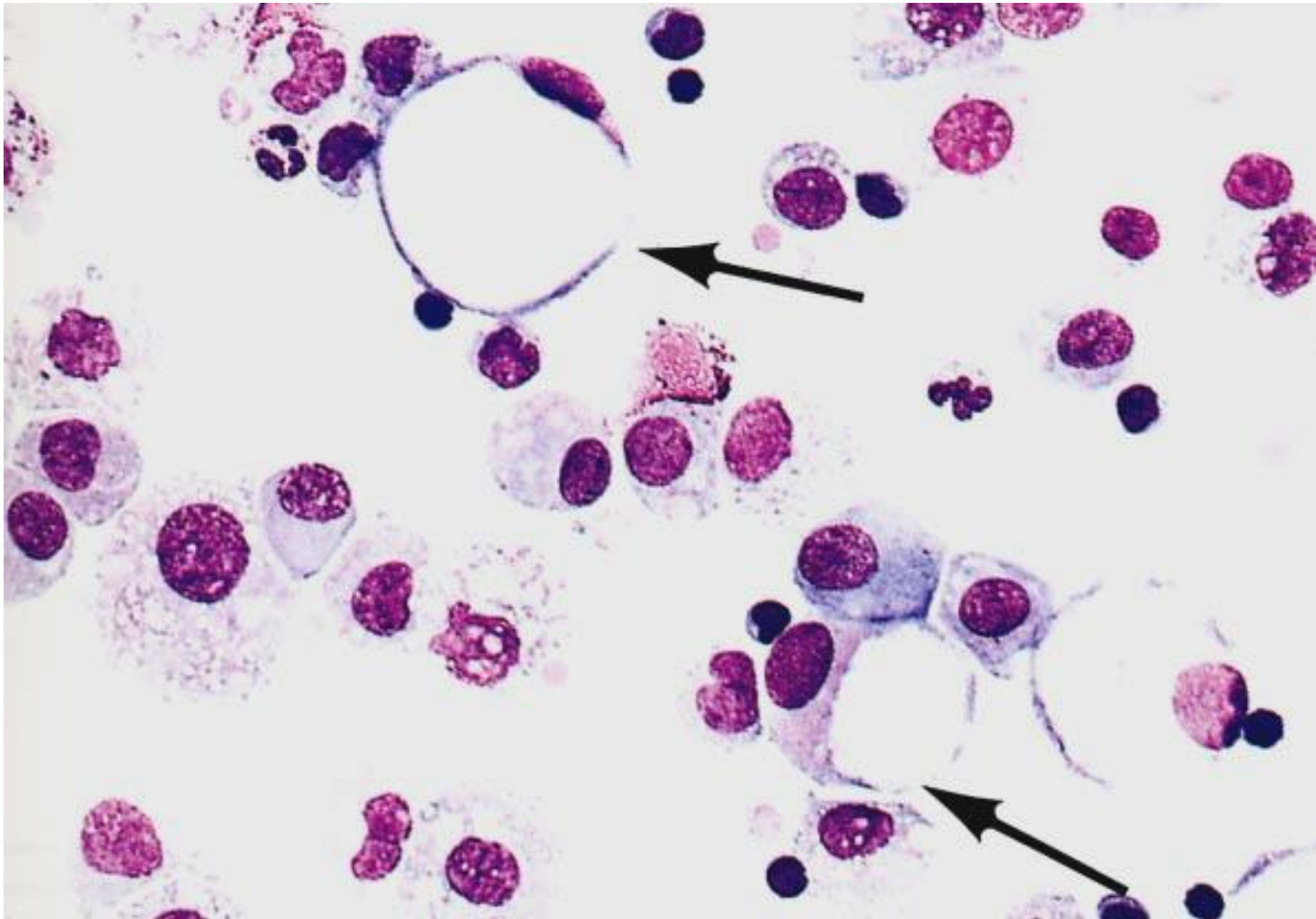
Female

Genital Tract	50.6 - 61%
Misc.	13.2 - 15.5%
Breast	10.3 - 17.6%
GI Tract	9.0 - 9.7%
Unknown	3.8 - 8.3%
Lung	0 - 1.7%

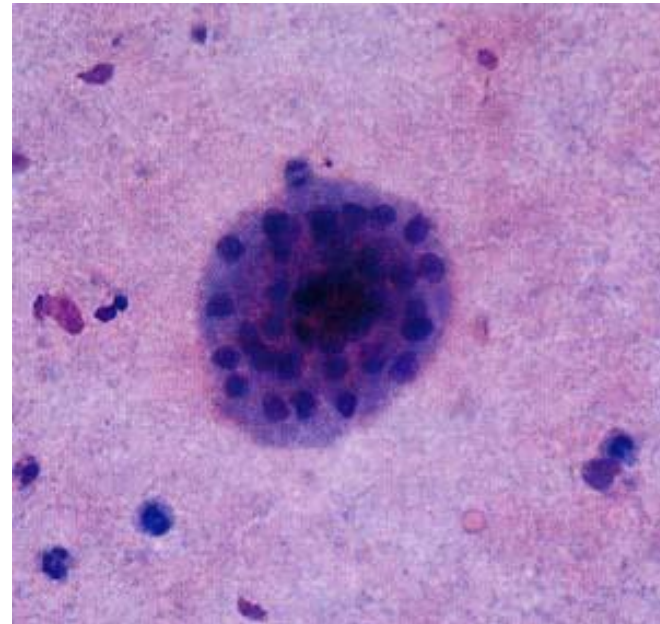
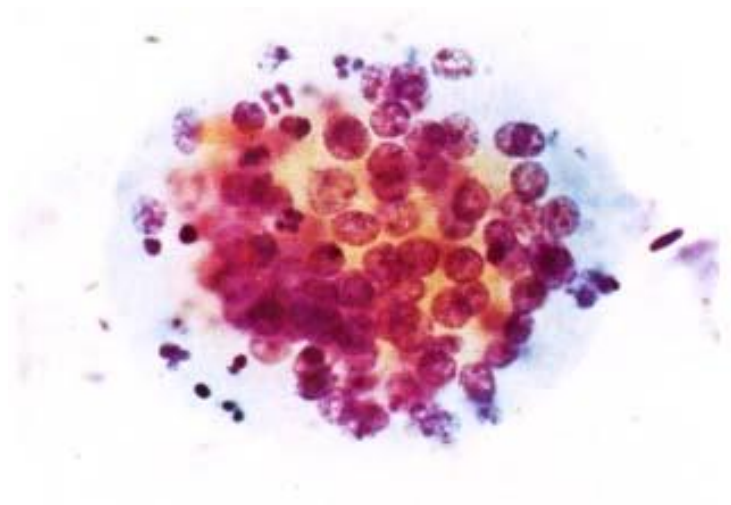
Macrophage & Mesothelial Cells

Macrophages

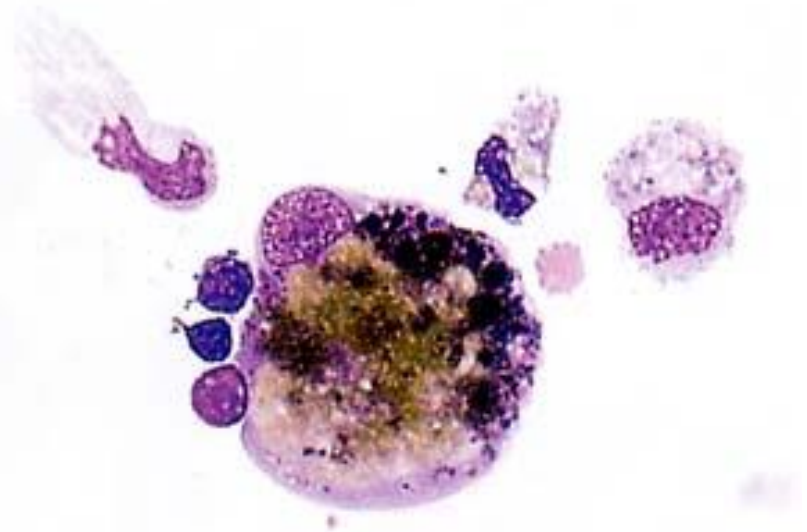
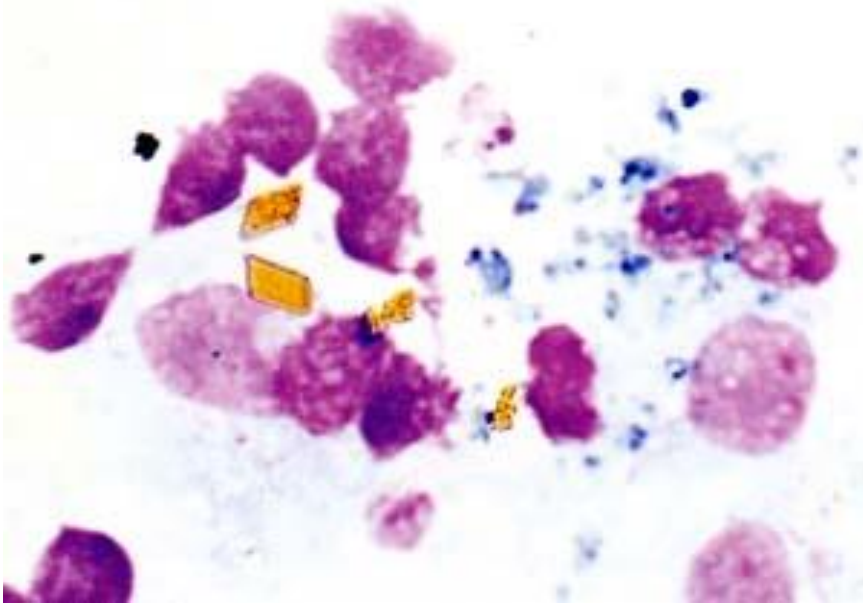


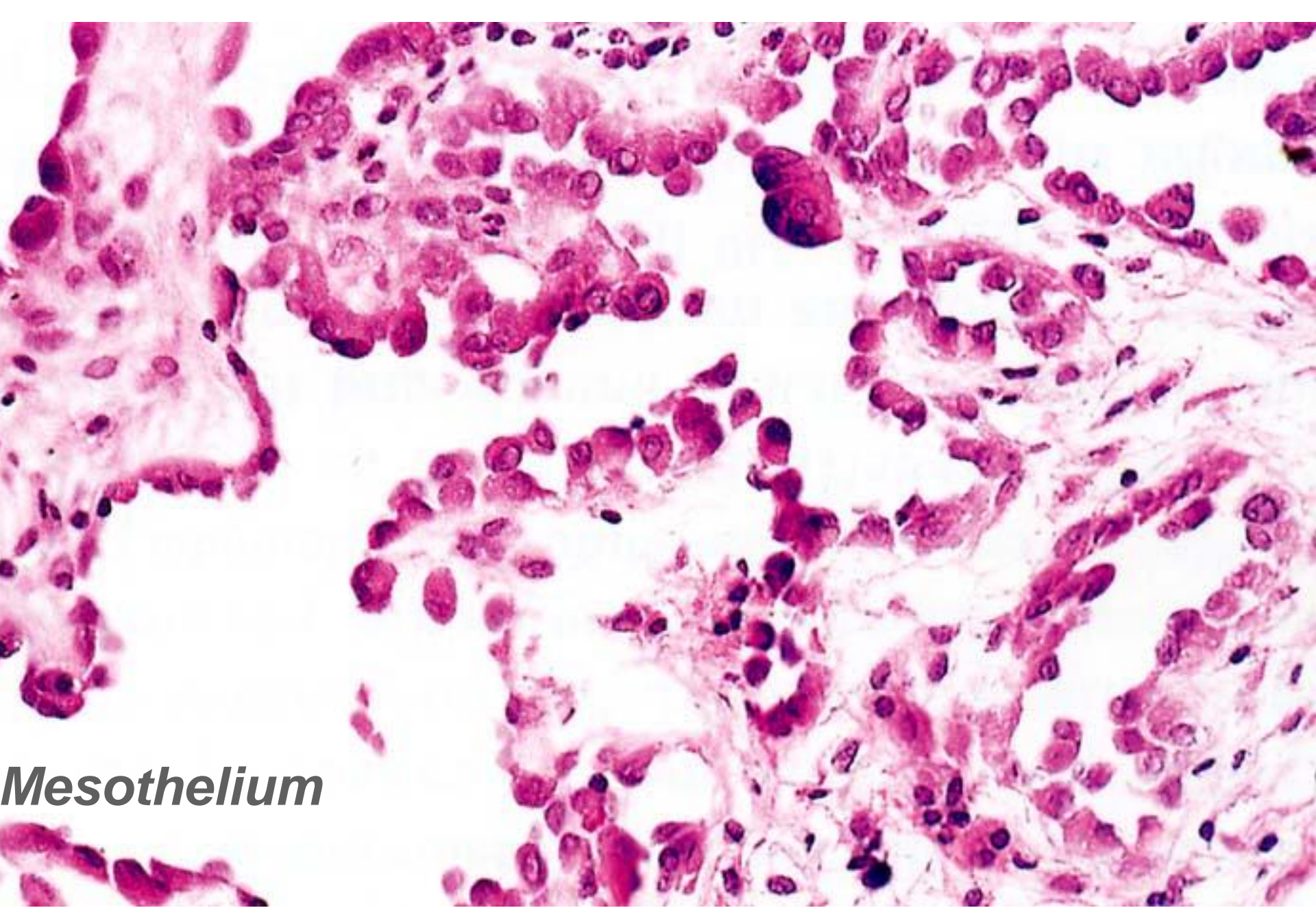


Multinucleated Giant Cells



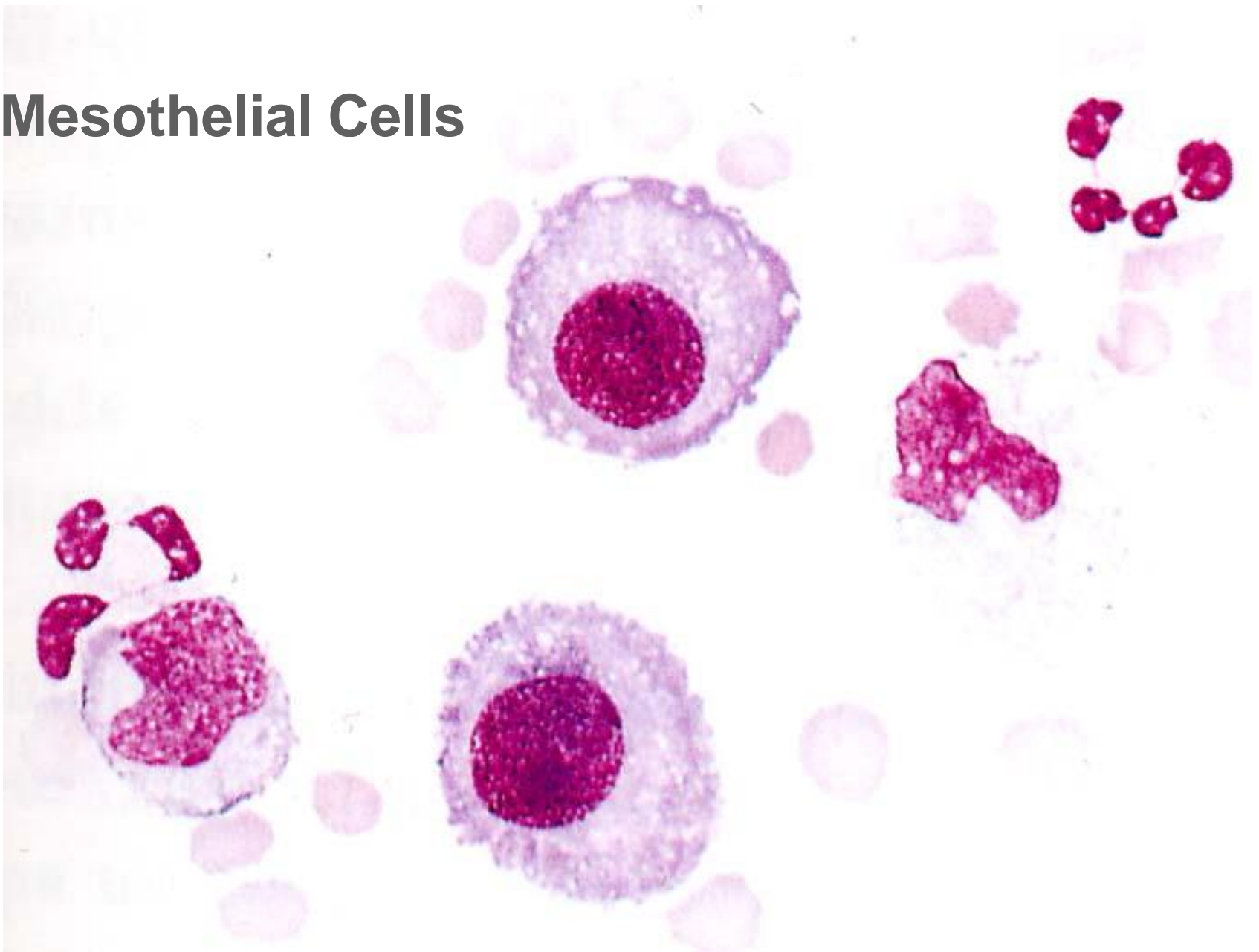
Pigment Laden Macrophages



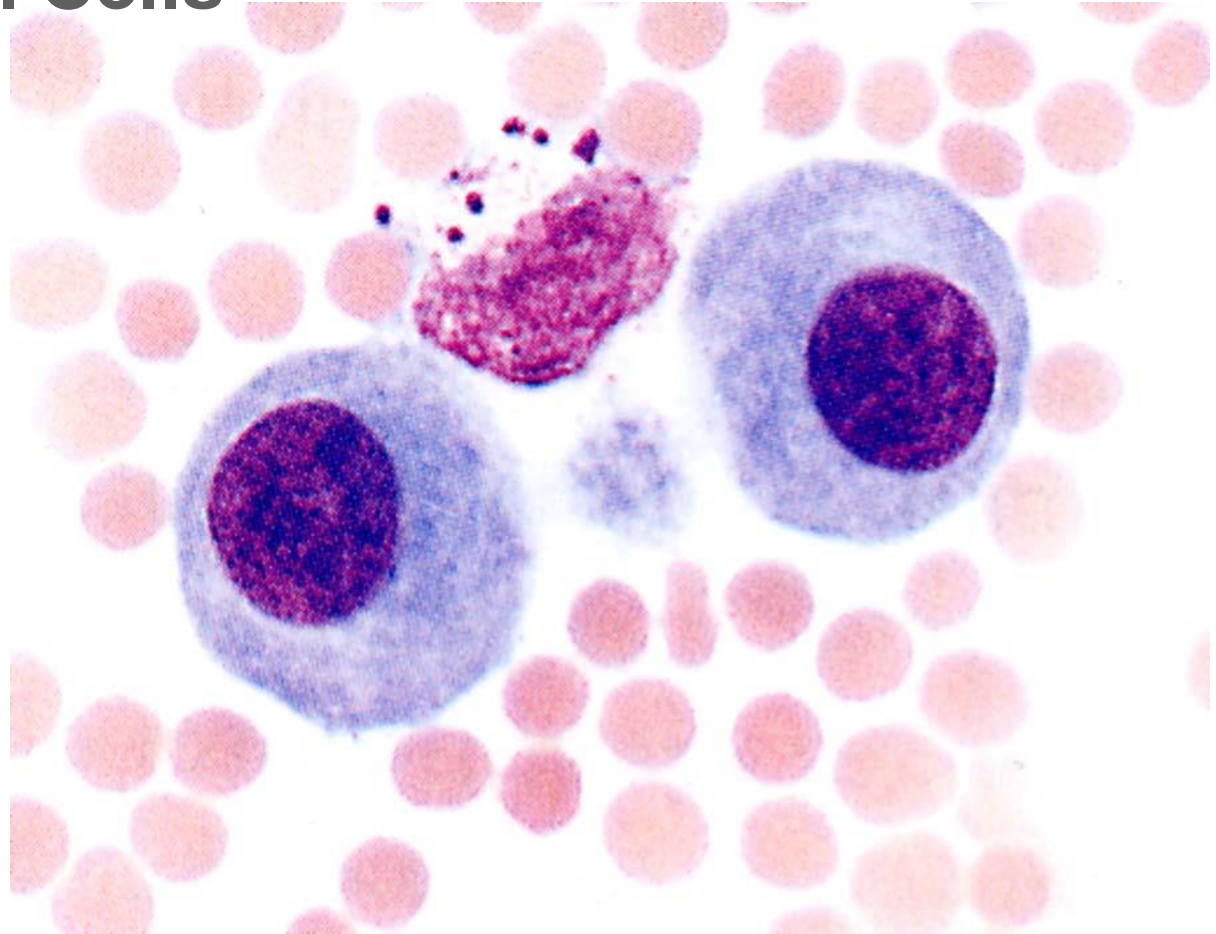


Mesothelium

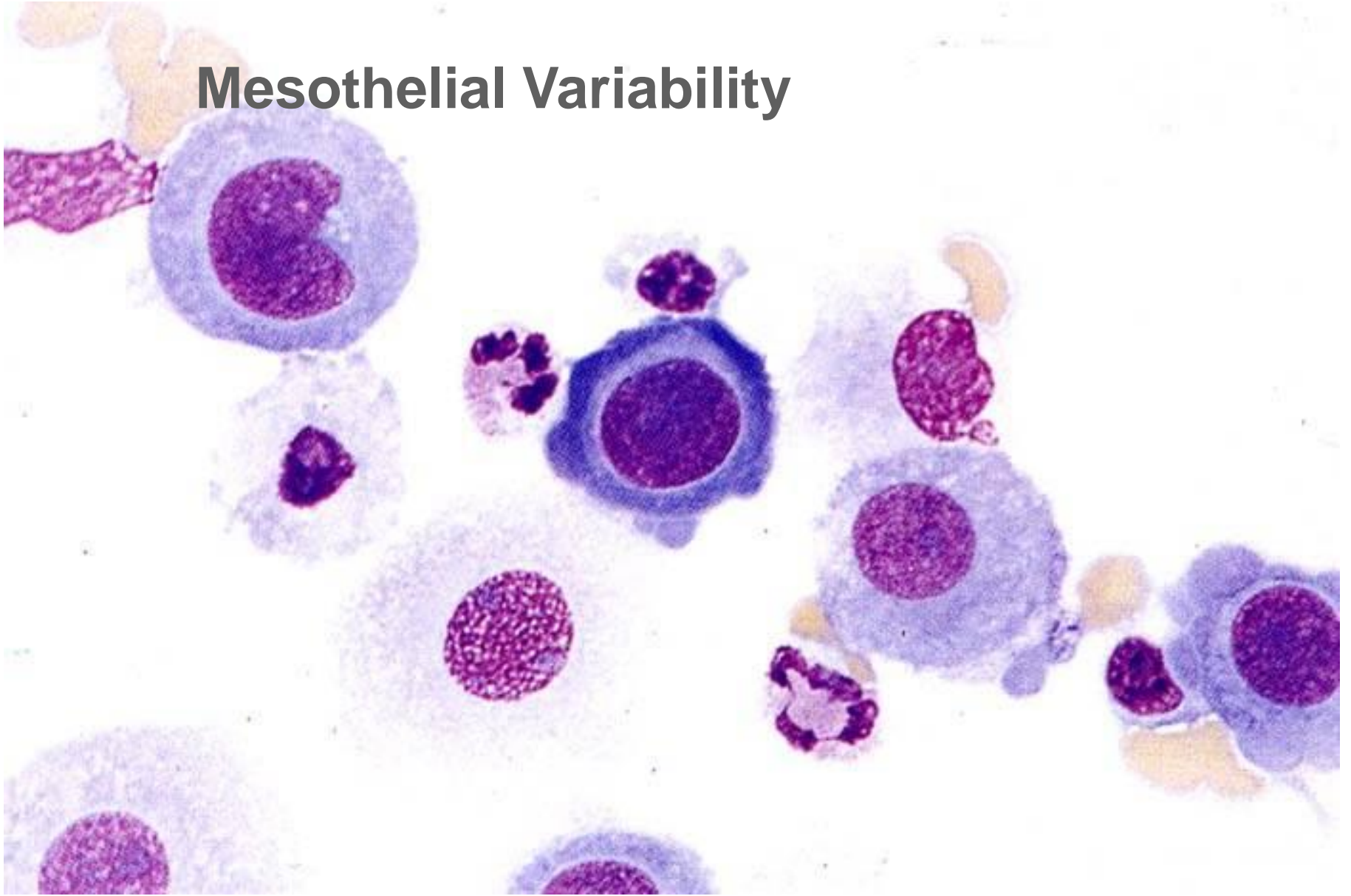
Mesothelial Cells



Mesothelial Cells



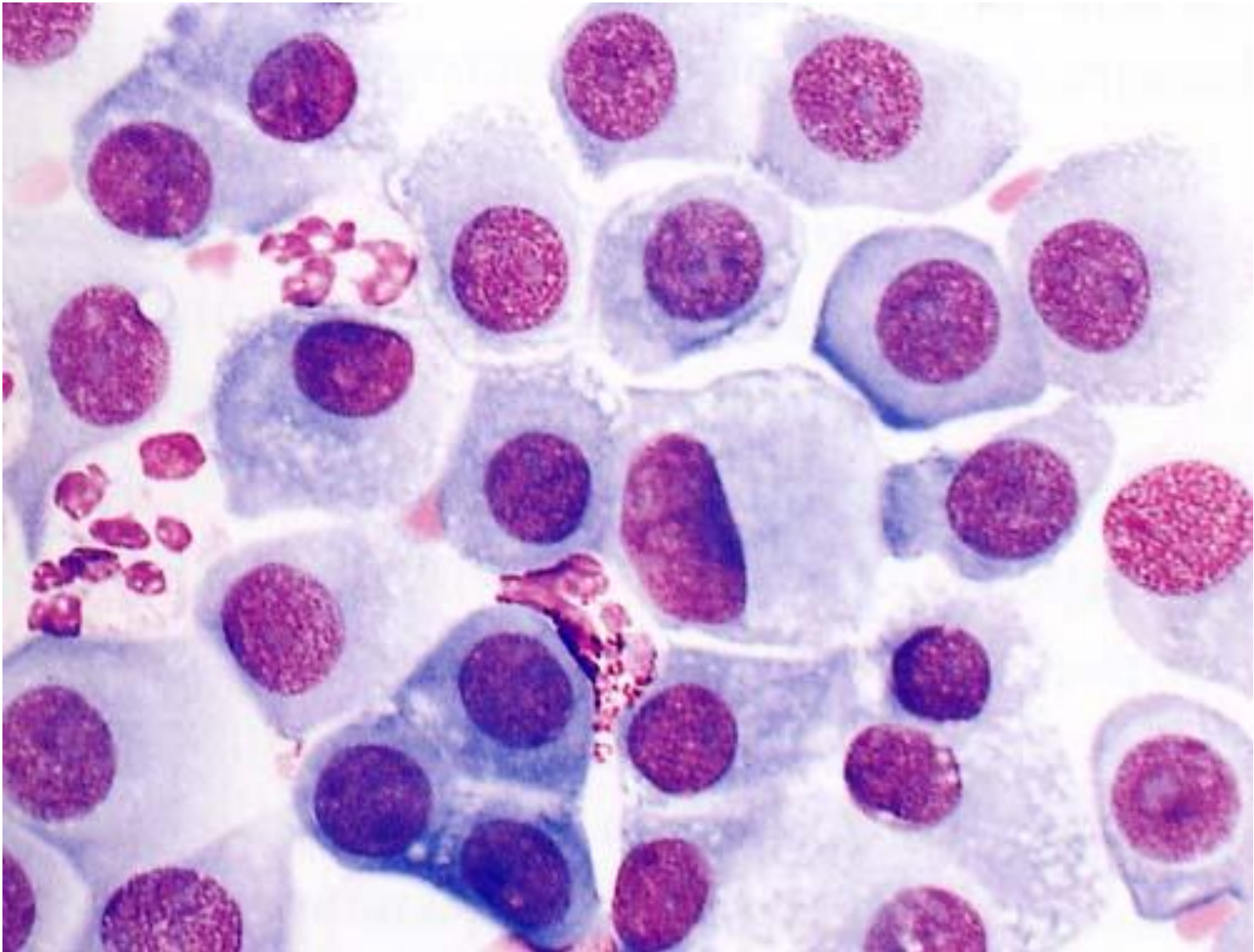
Mesothelial Variability



Signet Ring Mesothelials

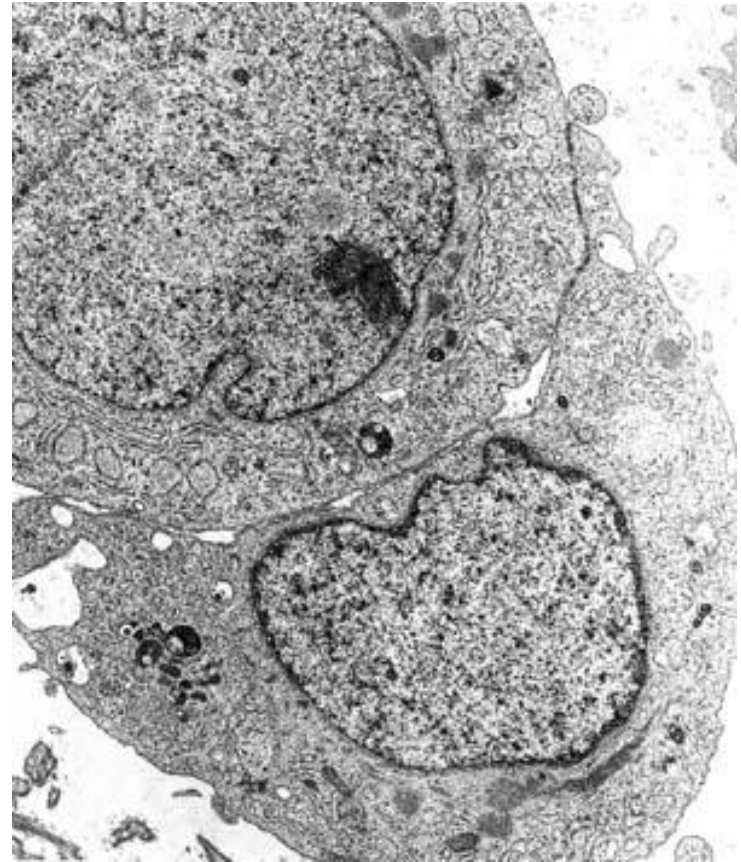
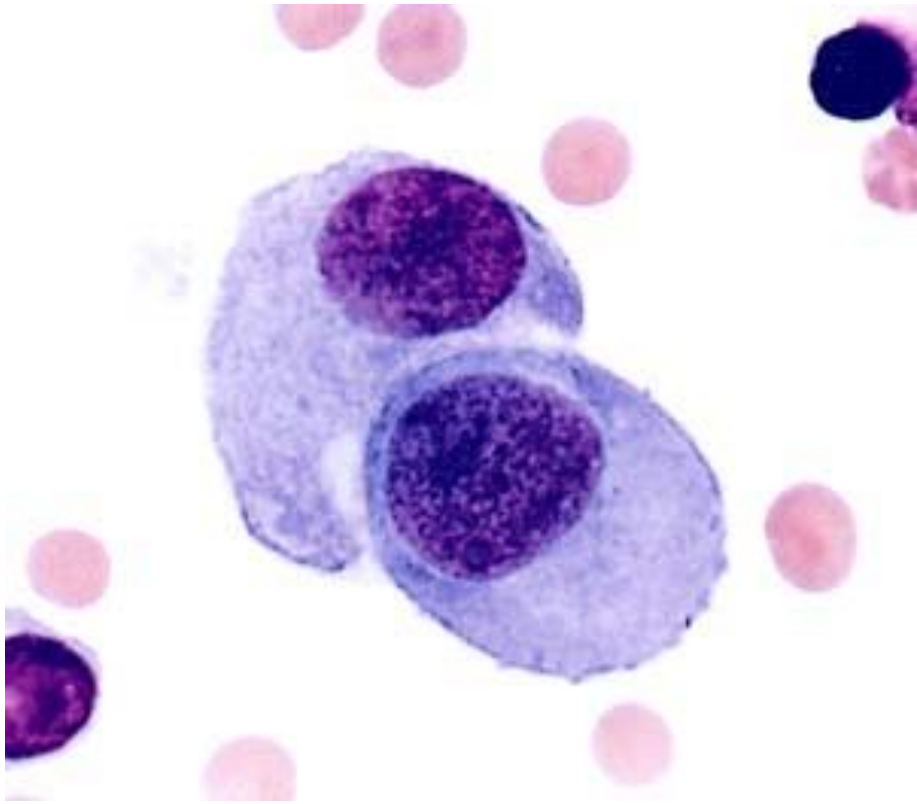


Mesothelial Windows

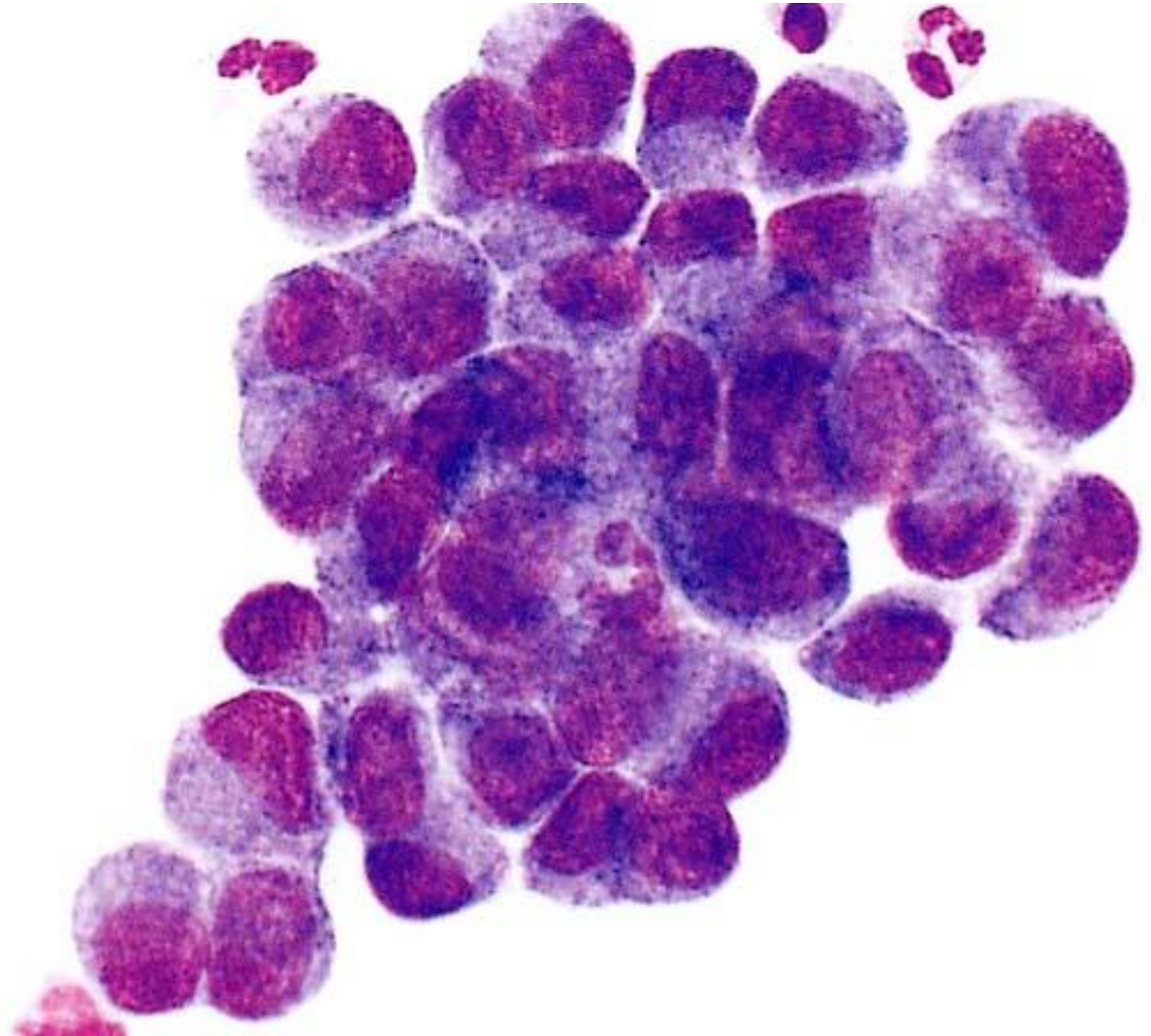


Mesothelial Hyperplasia

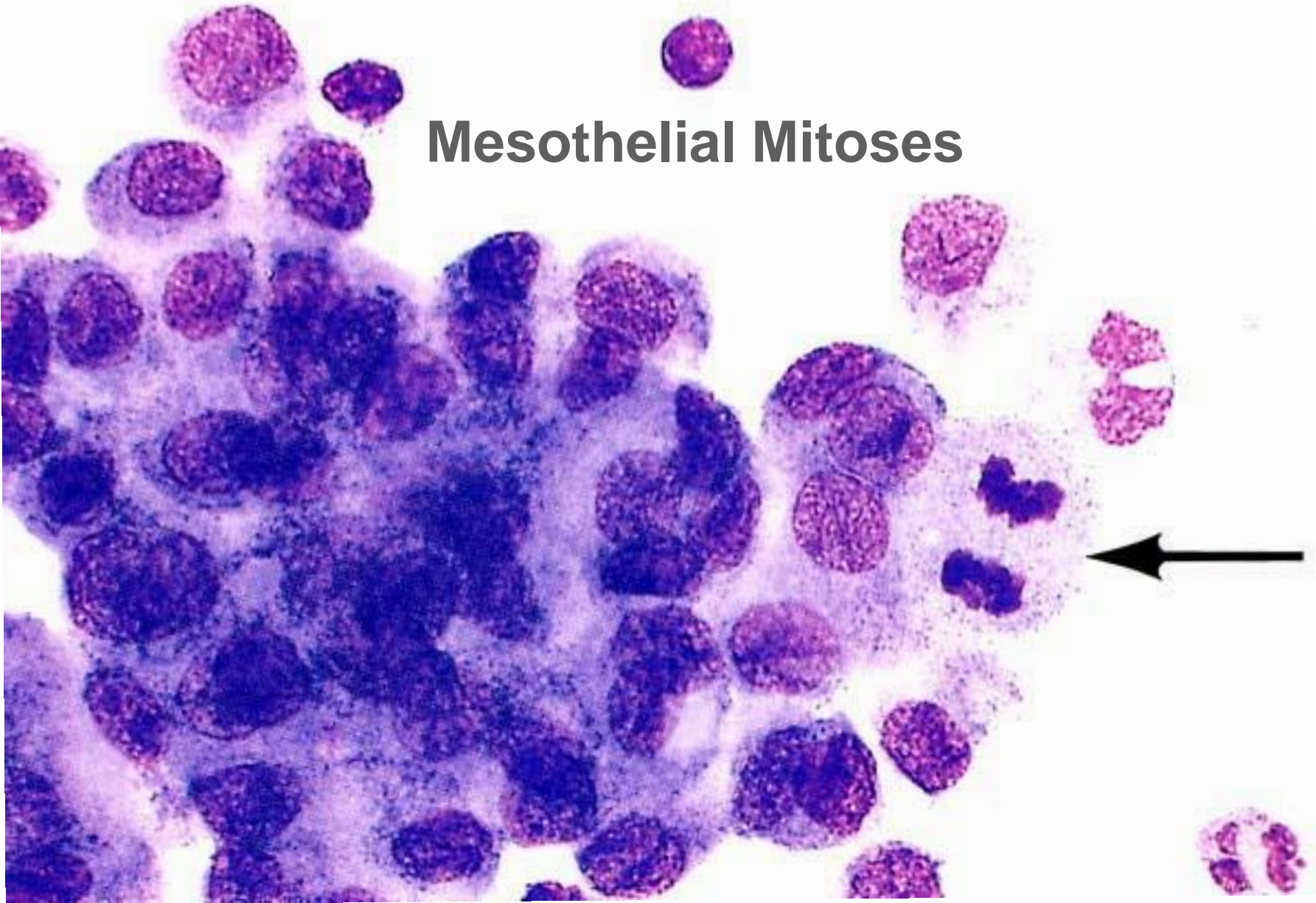
“Embracing” Mesothelials



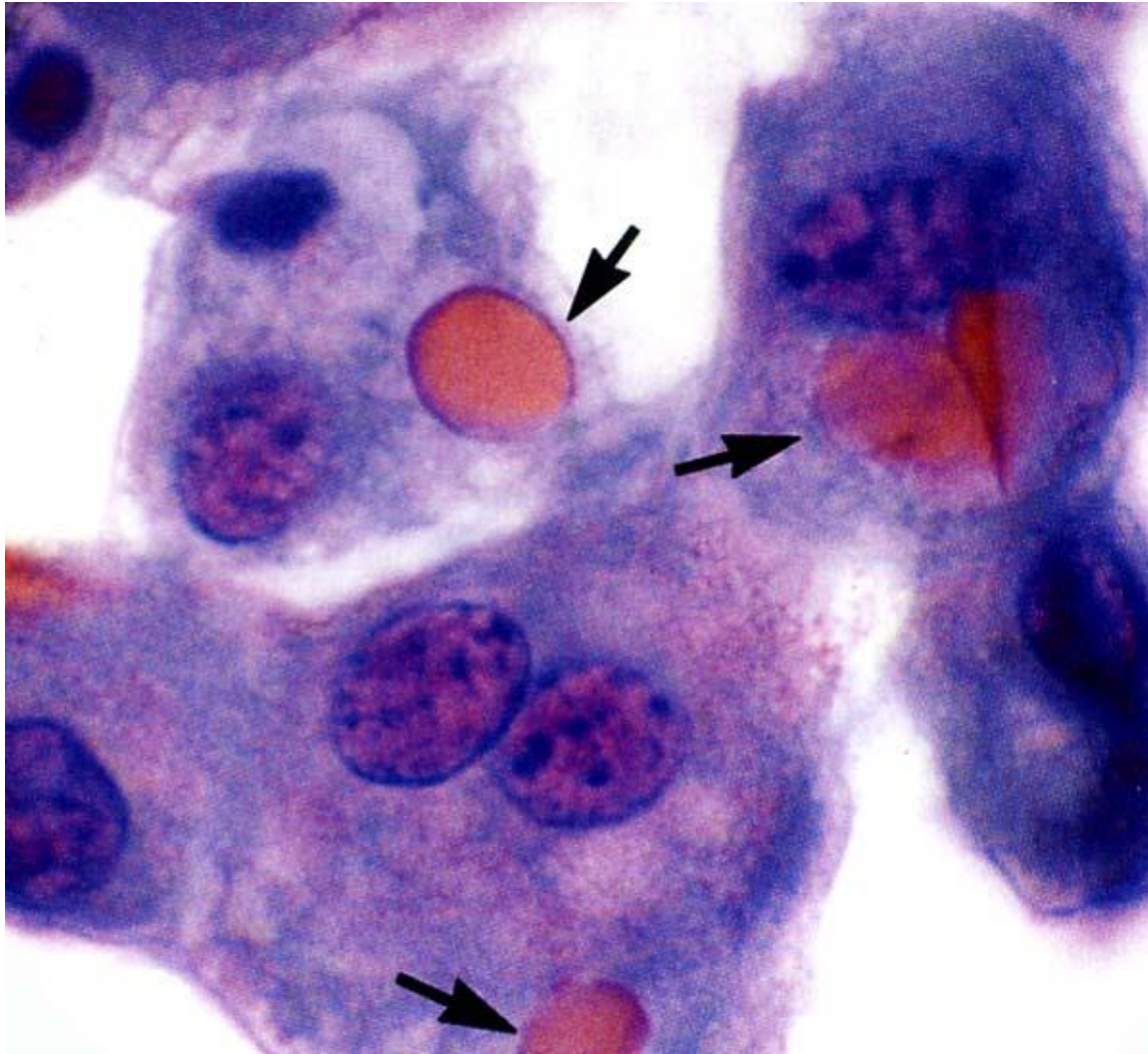
Mesothelial Clumps



Mesothelial Mitoses

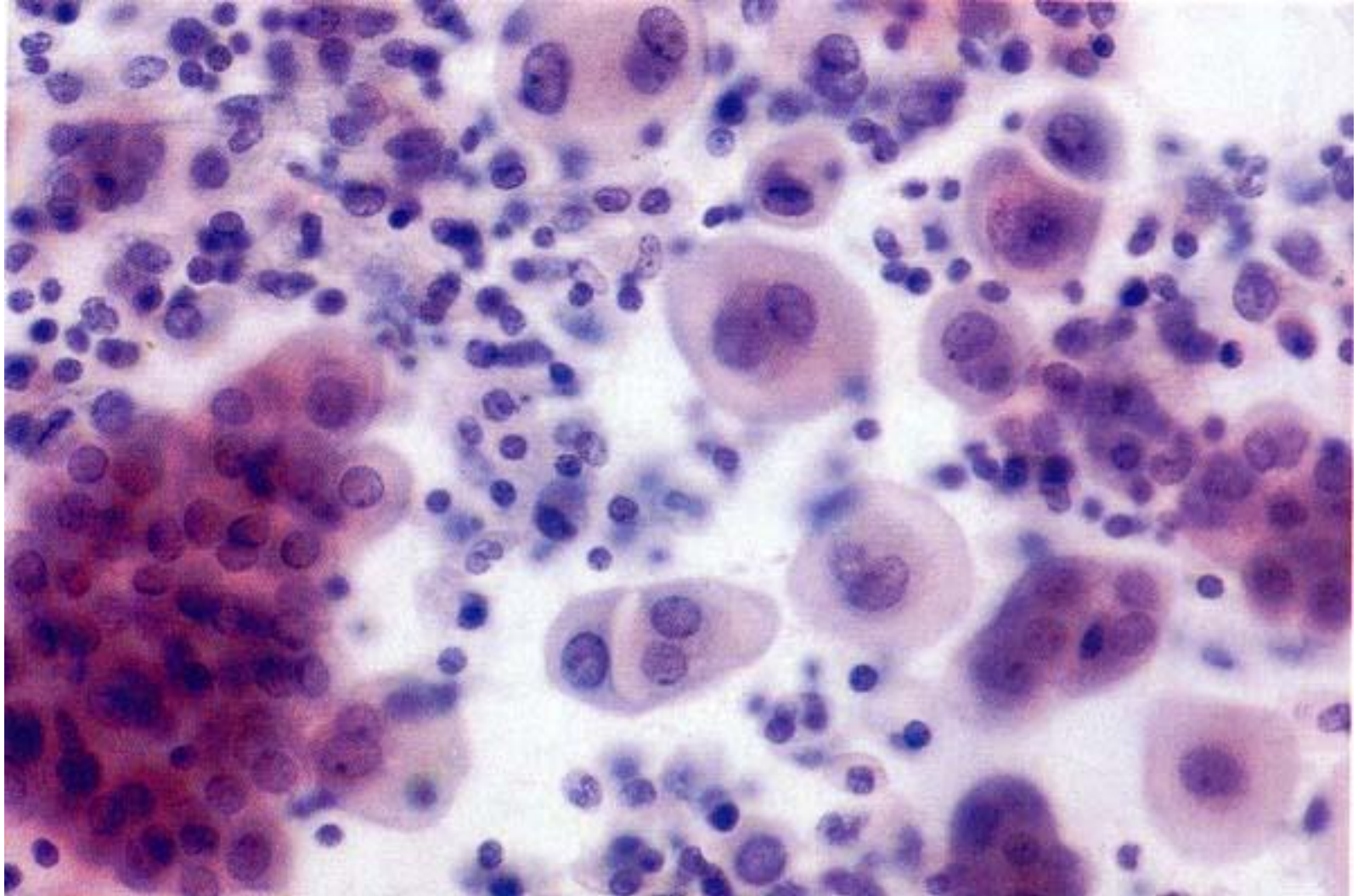


Mesothelial erythrophagocytosis

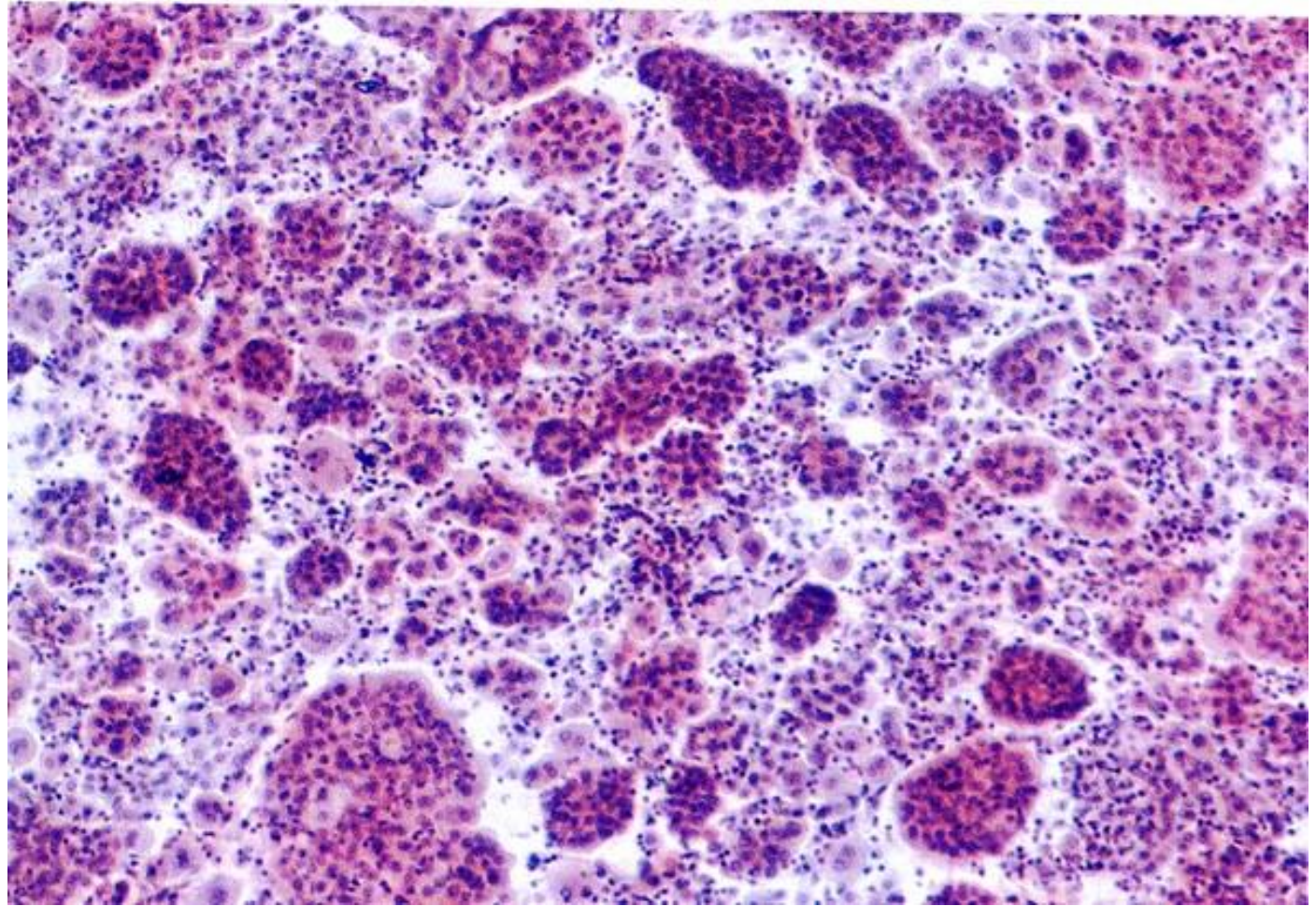


**Pap
stain**

Diagnosis?



Malignant Mesothelioma



Immunohistochemistry

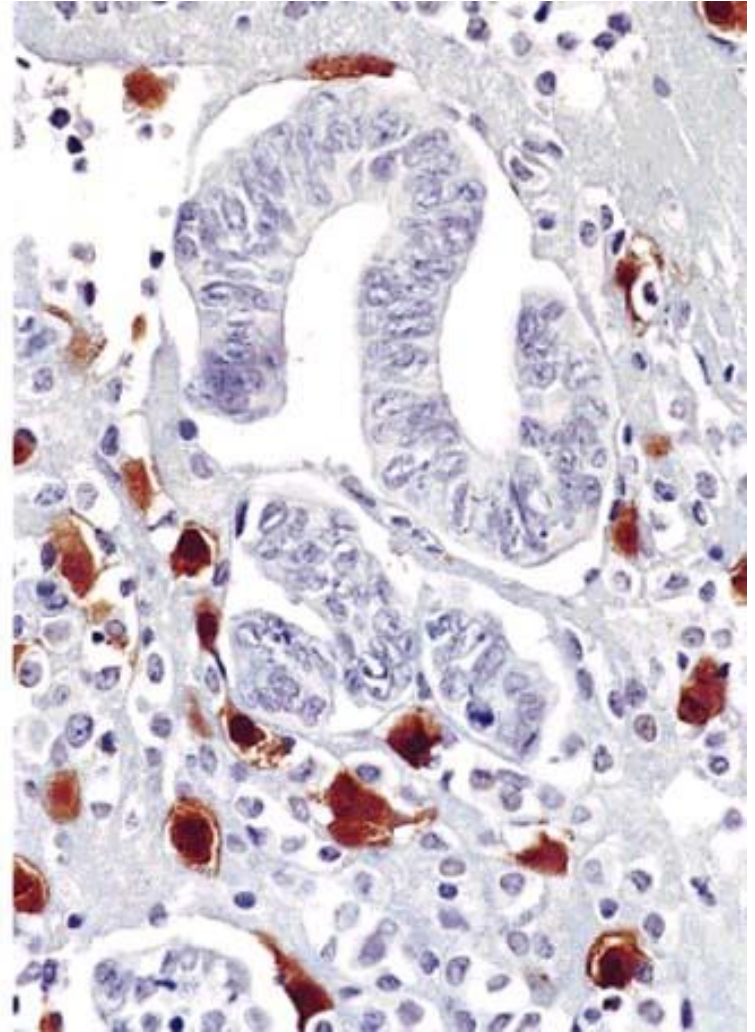
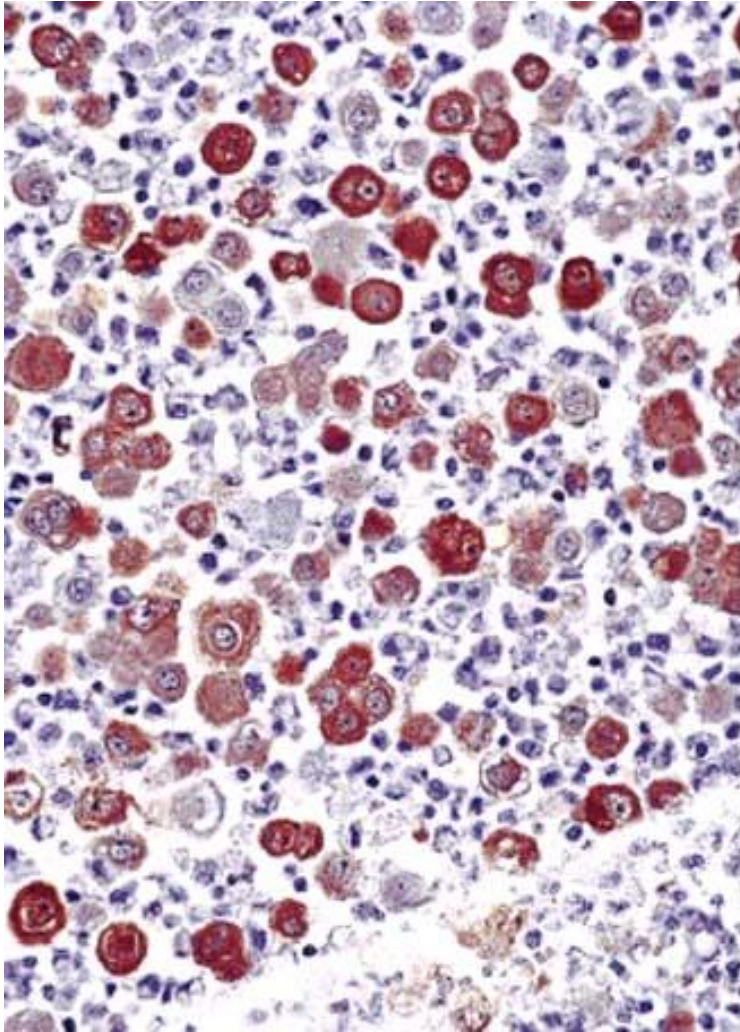
Mesothelioma

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

Adenocarcinoma

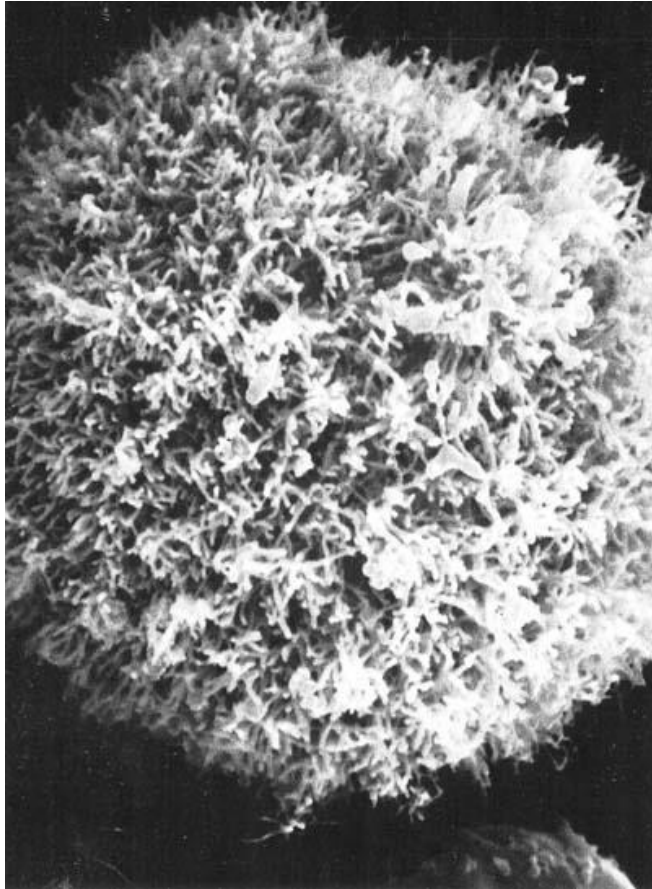
- ✓ **CEA**
- ✓ **CD15**
- ✓ **Ber EP-4**
- ✓ **B72.3**
- ✓ **Homogeneously distributed stain**

Calretinin immunostain

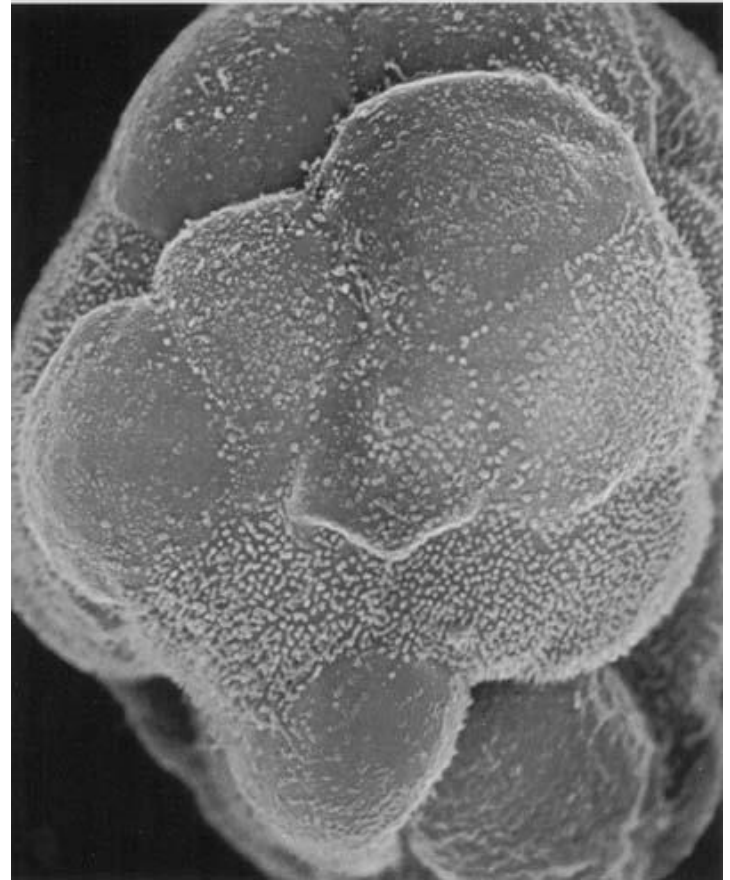


Electron Microscopy

Mesothelioma



Adenocarcinoma

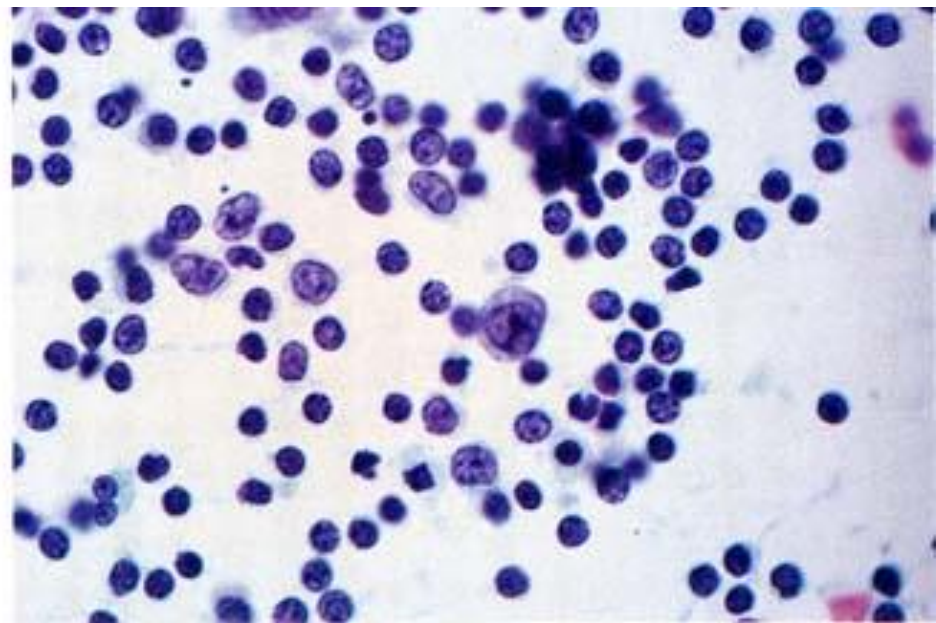
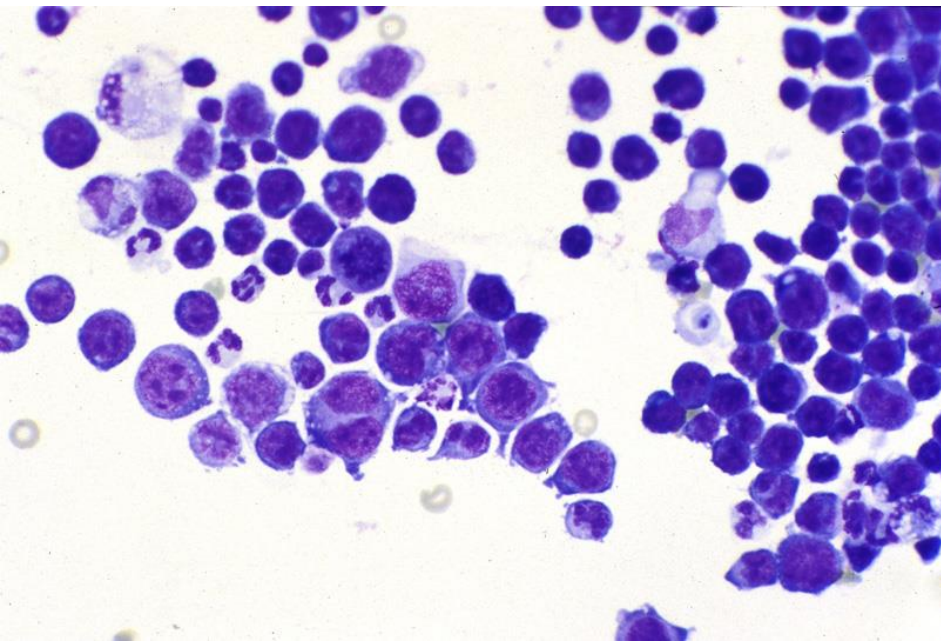


Benign vs. Malignant

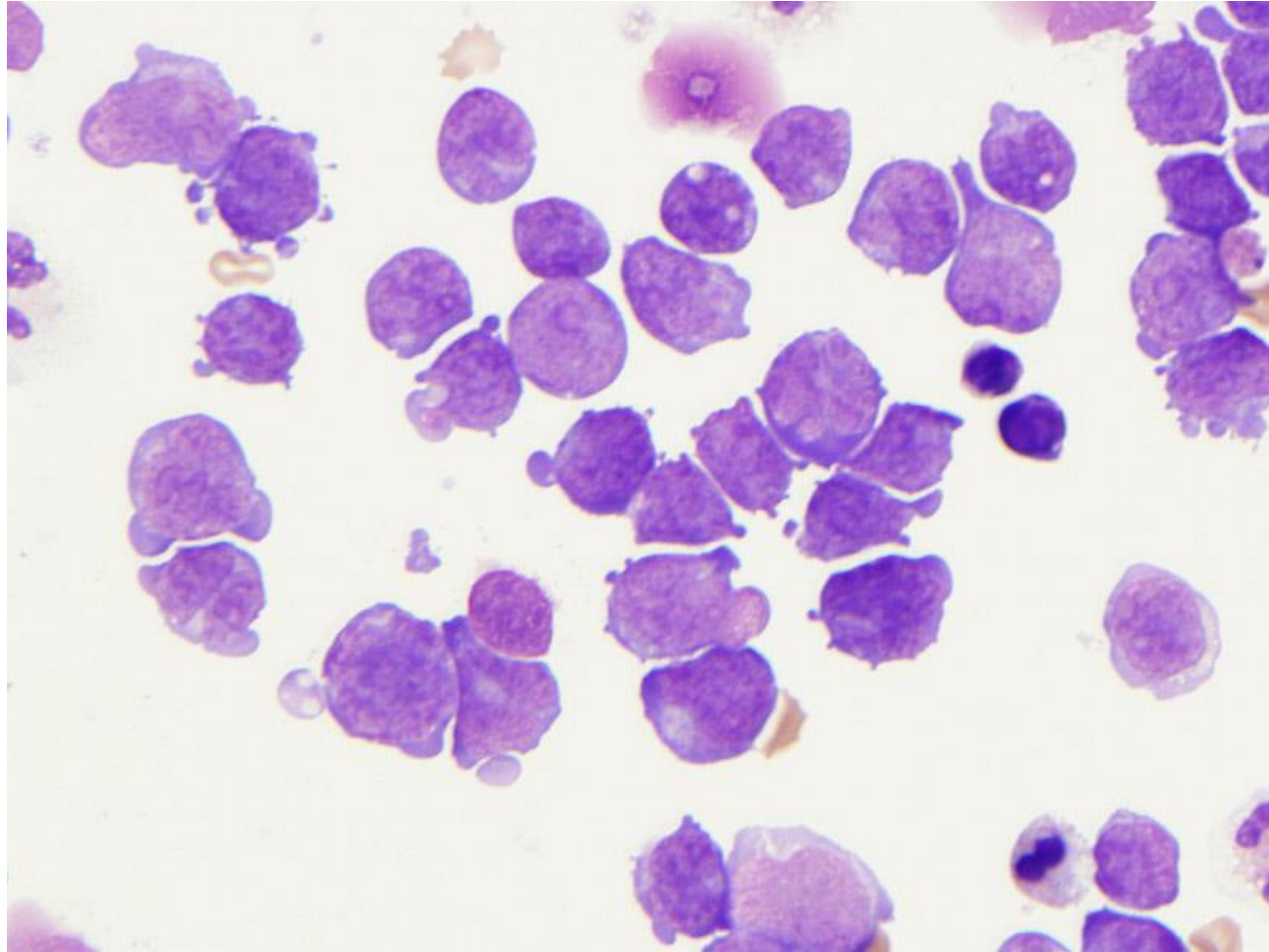
Unusual homogenous population

- ***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

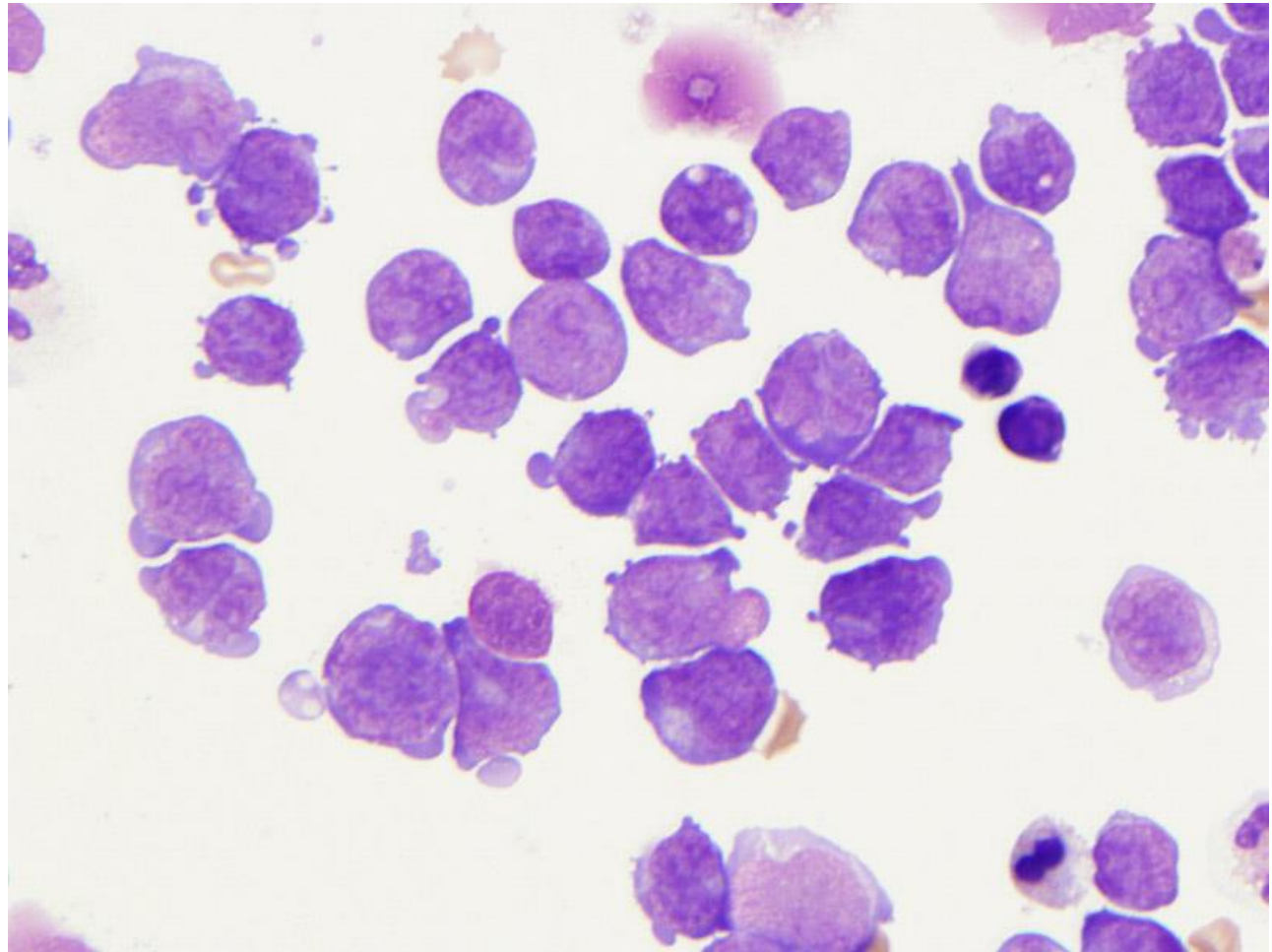
Lymphocytes



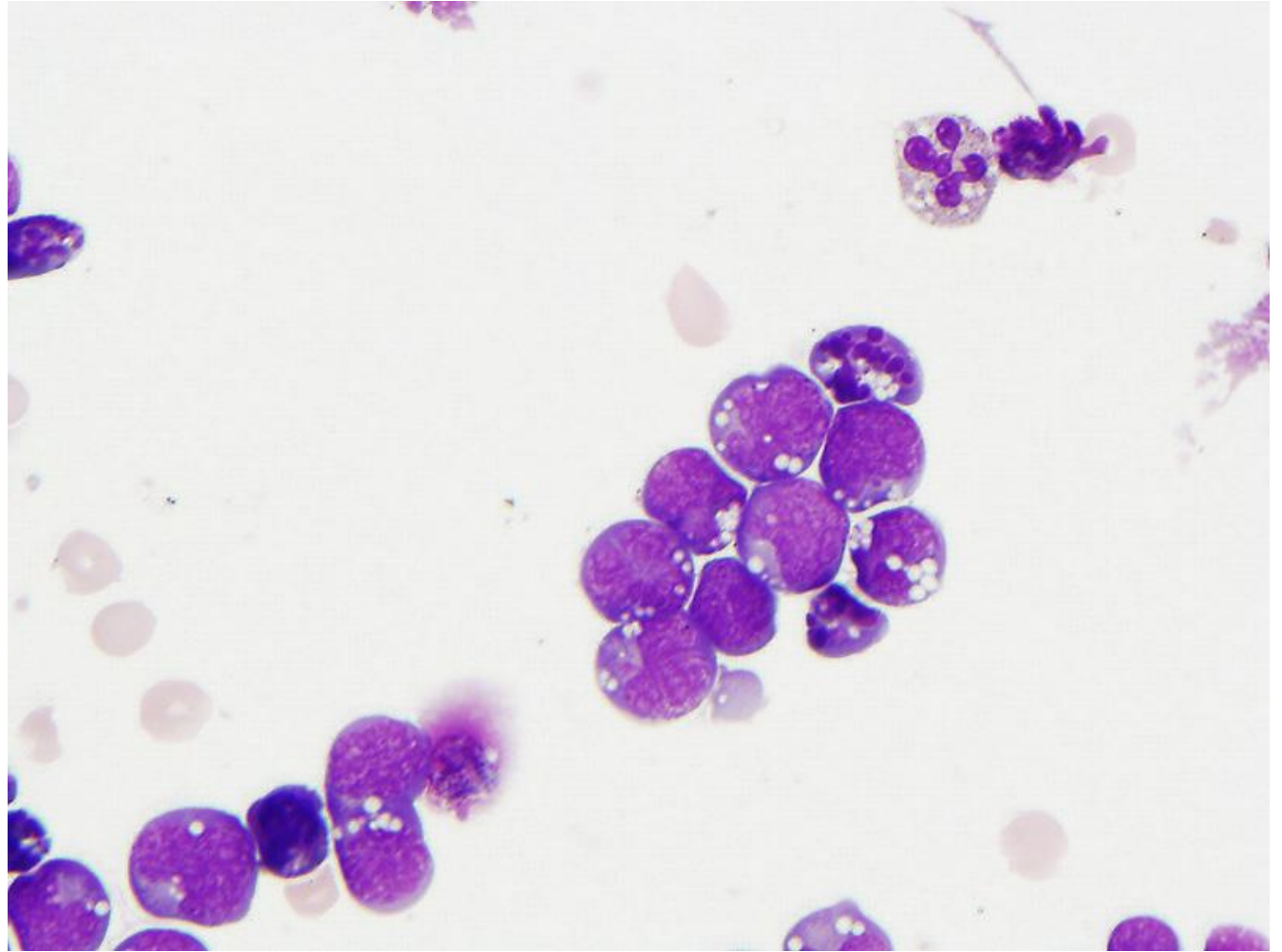
Diagnosis?



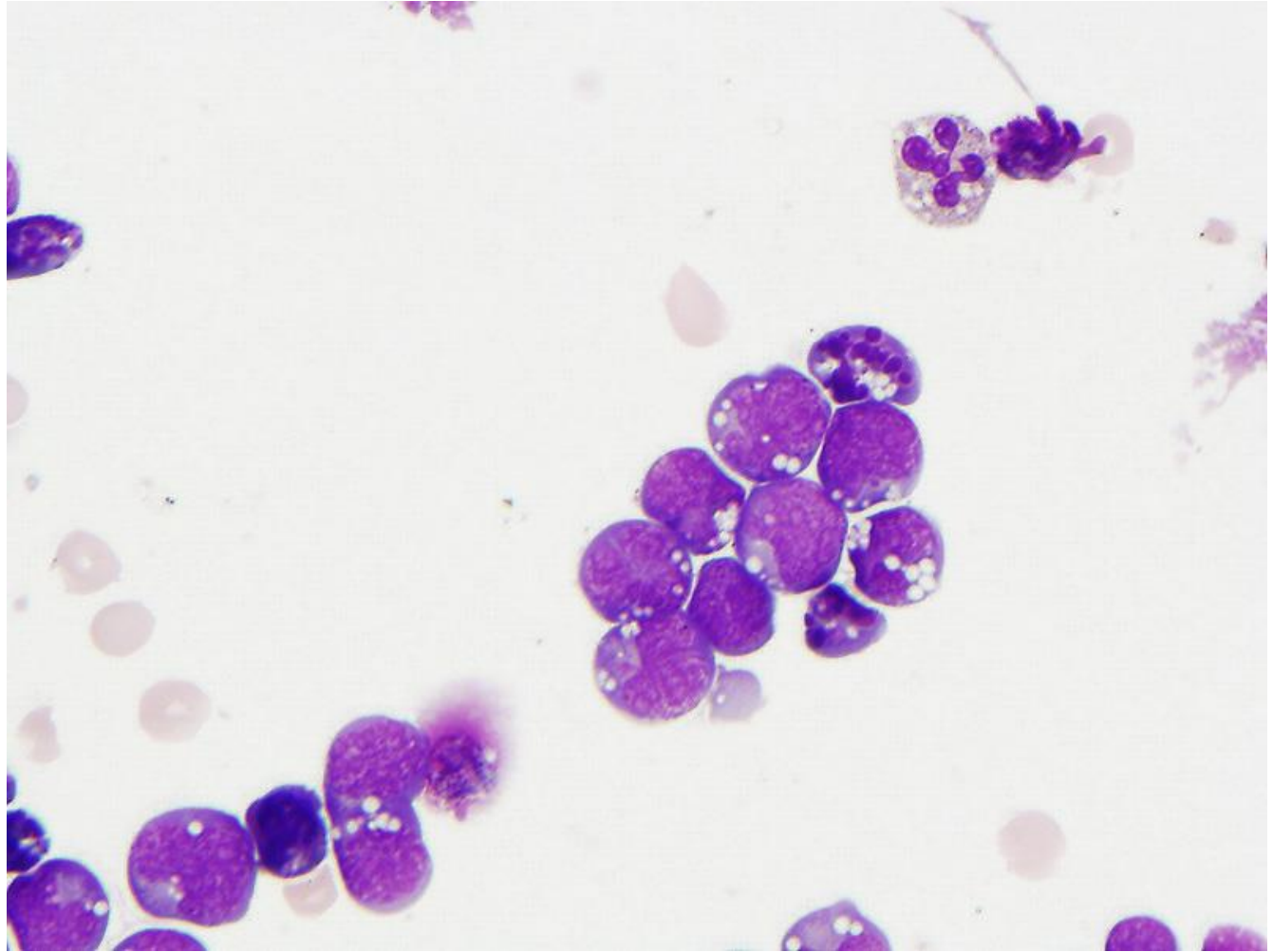
Large cell lymphoma



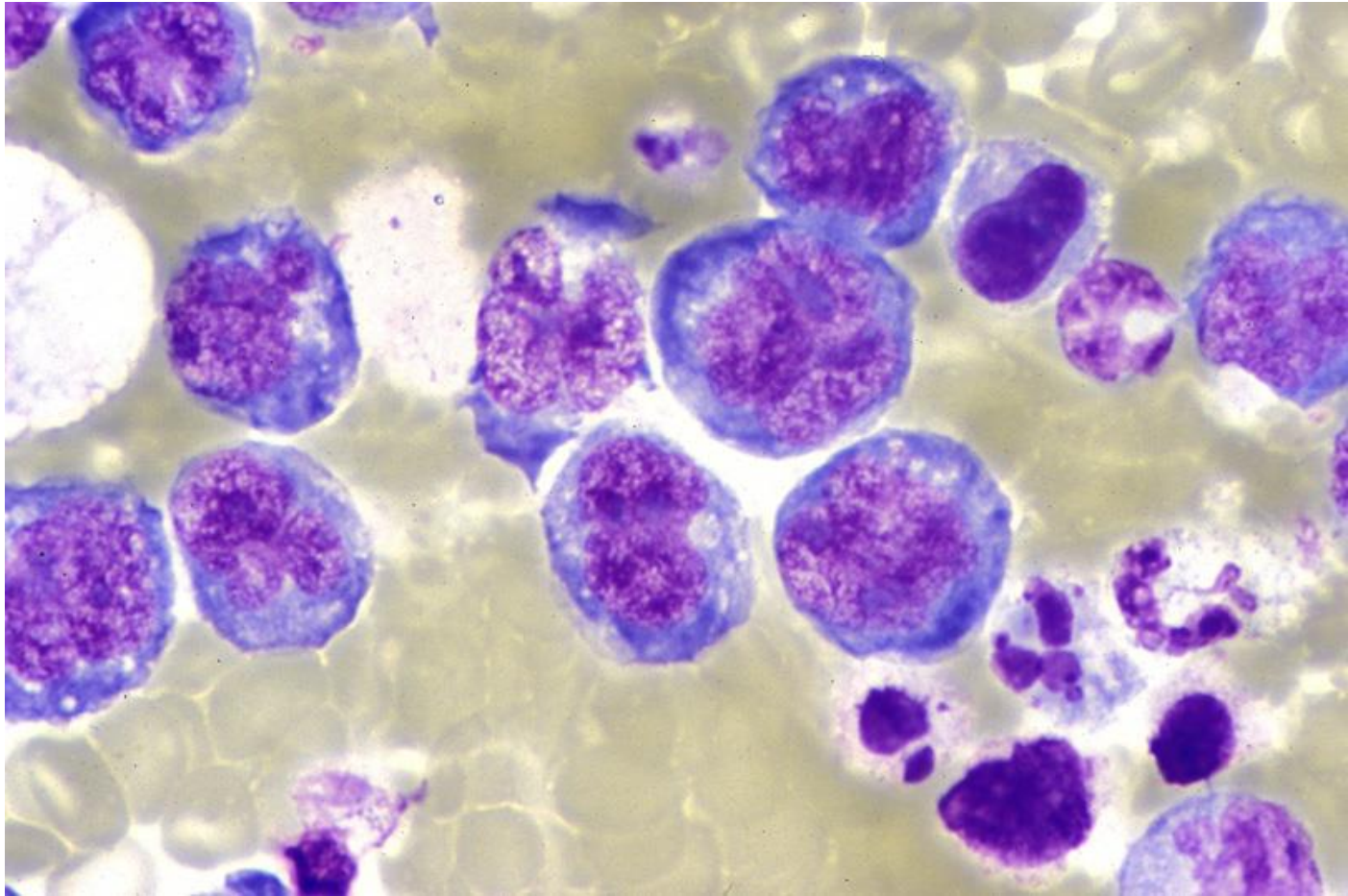
Diagnosis?



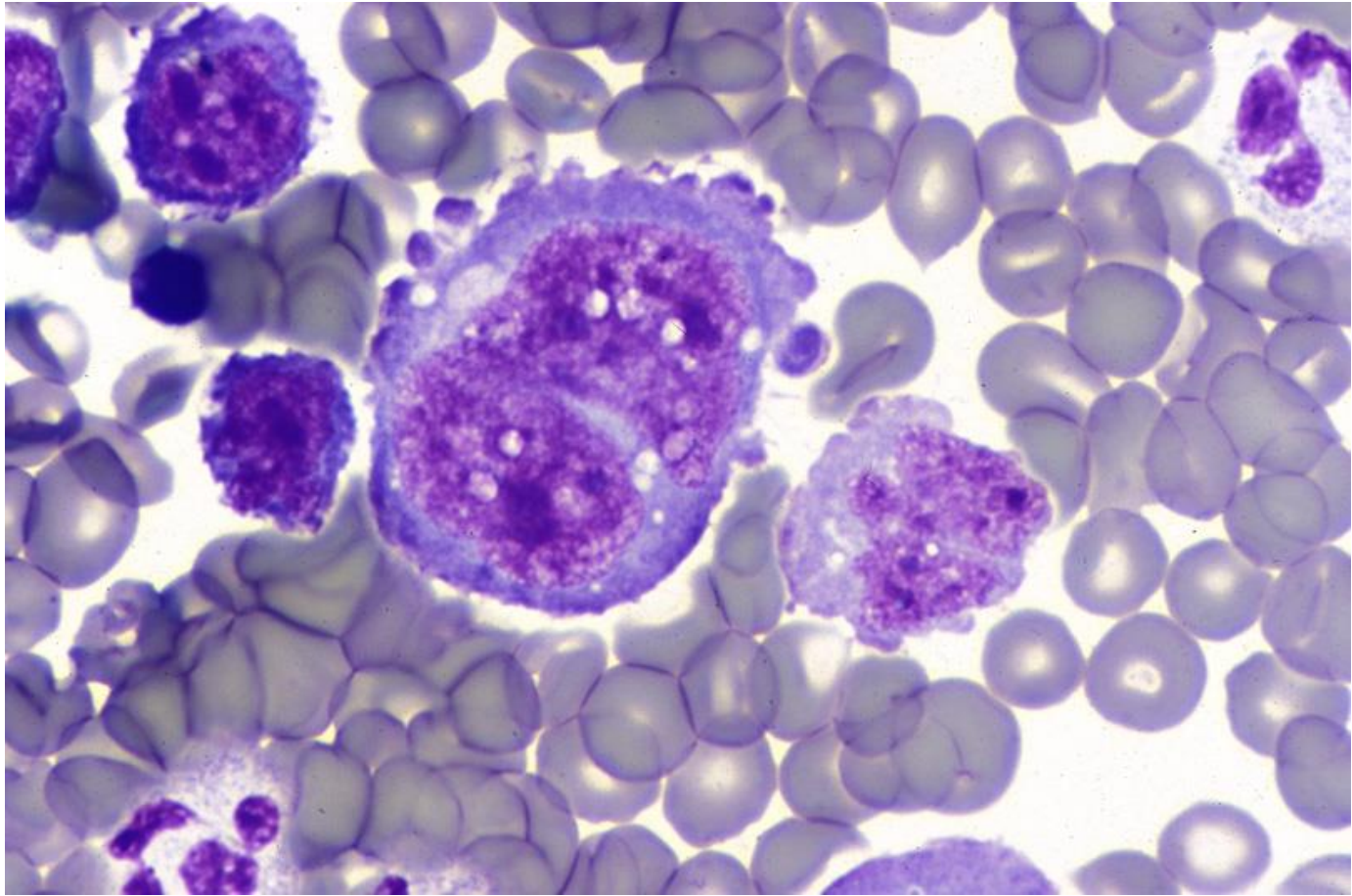
Burkitt lymphoma



Diagnosis?



Primary Effusion Lymphoma



Features of Adenocarcinoma

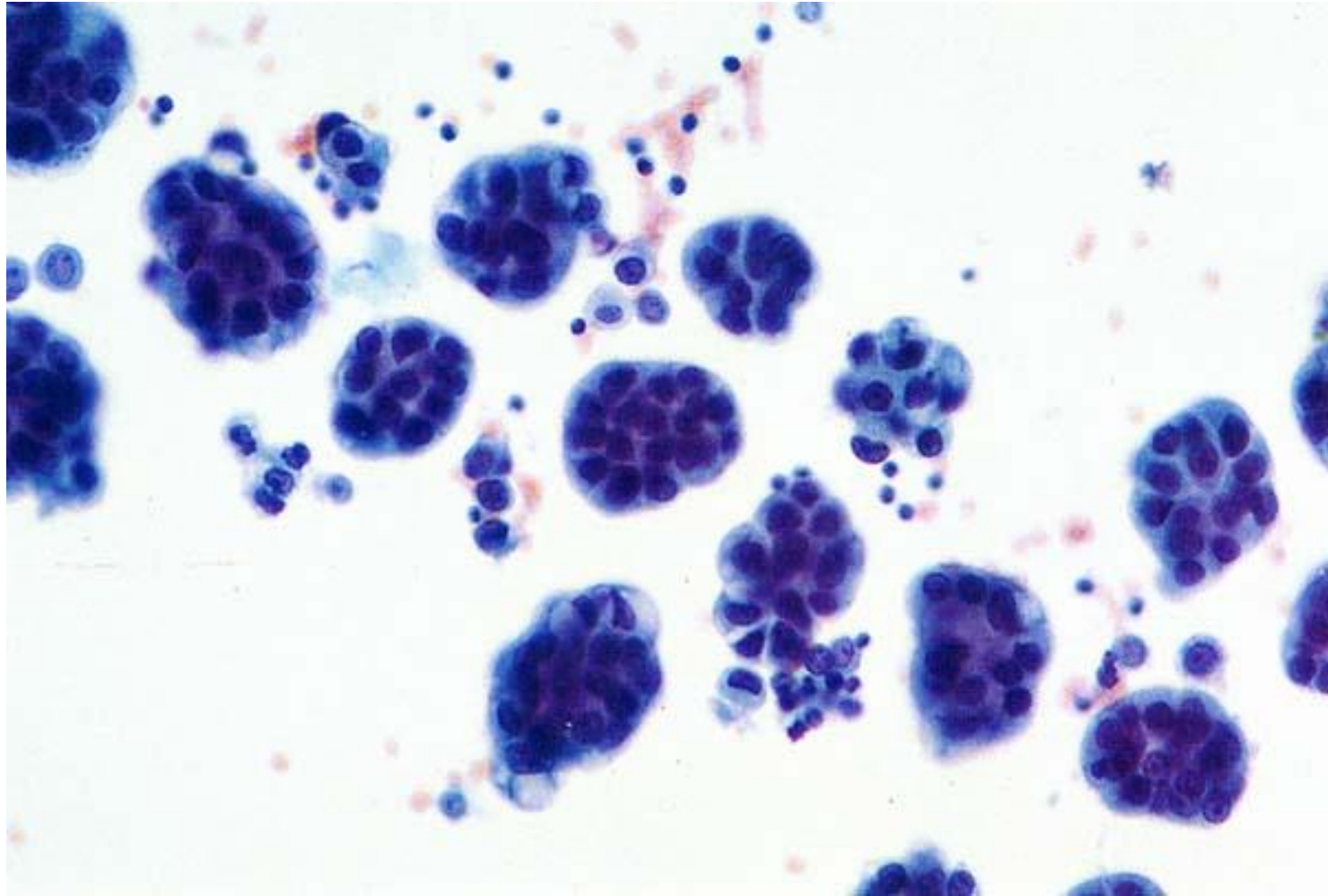
Cell groups

- ✓ **Solid cell balls**
- ✓ **Papillary forms**
- ✓ **Free-floating acini**
- ✓ **Columnar cell fronds**
- ✓ **Single-cell rows**

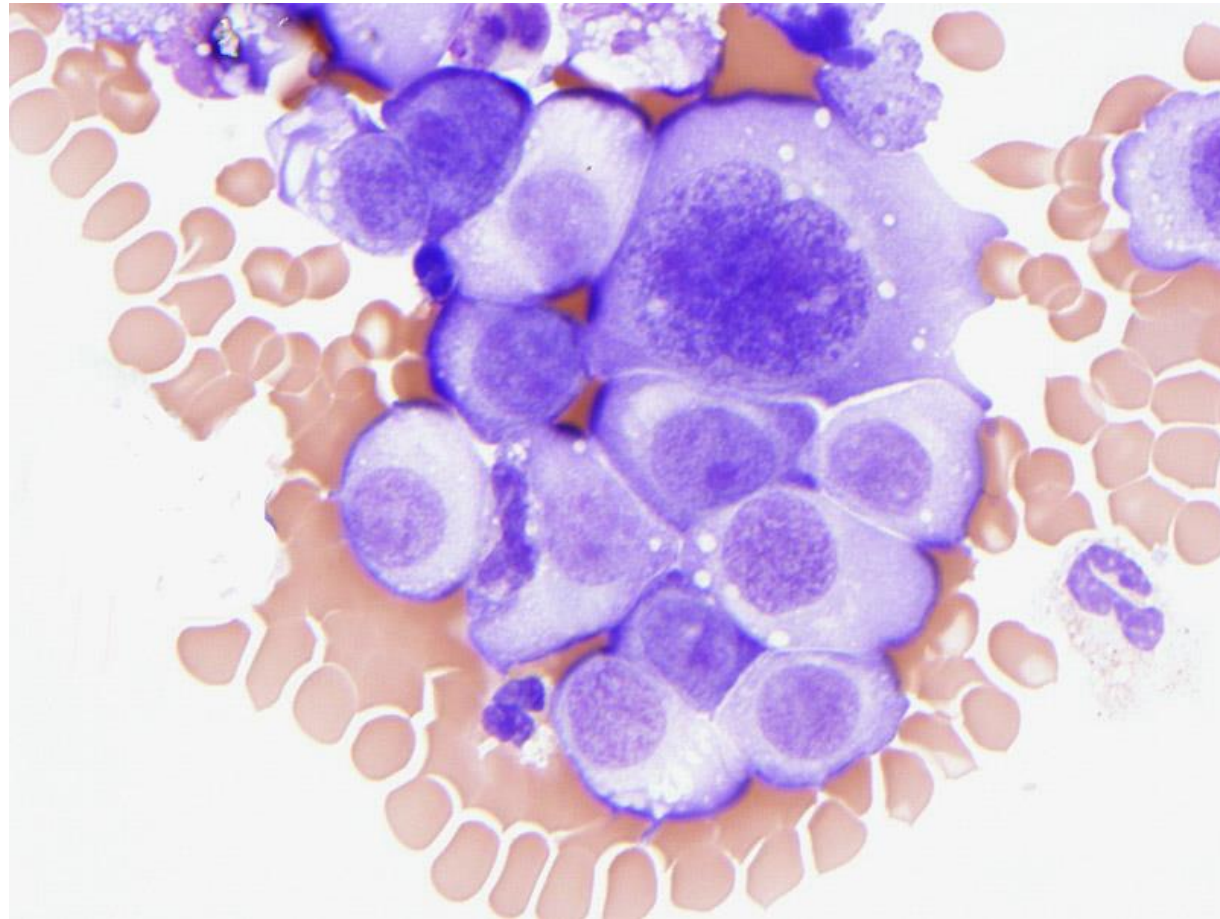
Individual cells

- ✓ **Signet-ring forms**
- ✓ **Intracytoplasmic lumina**
- ✓ **Clear cell change**
- ✓ **Intracytoplasmic dot**
- ✓ **Multinucleated giant cells**

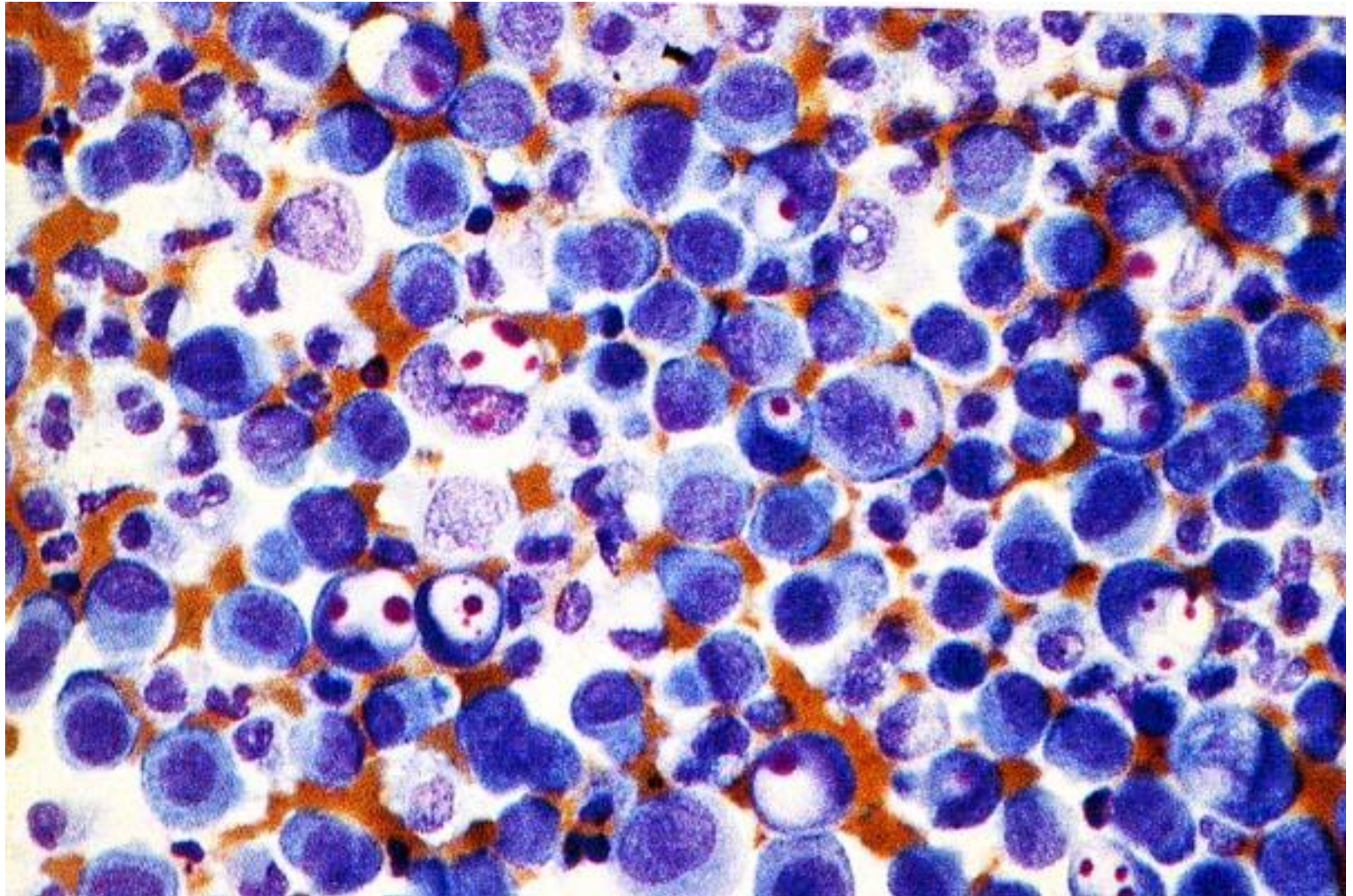
Breast carcinoma



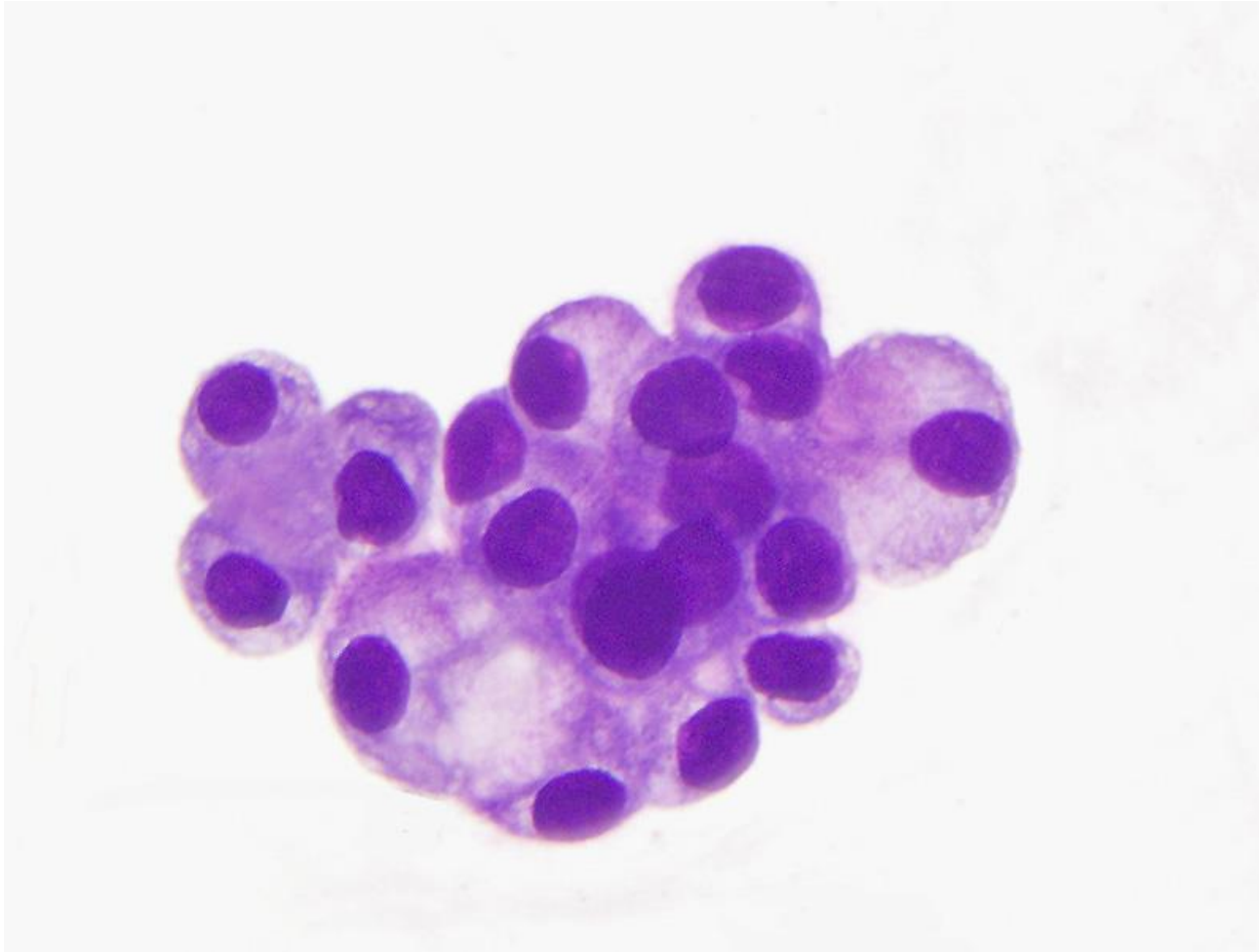
Diagnosis?



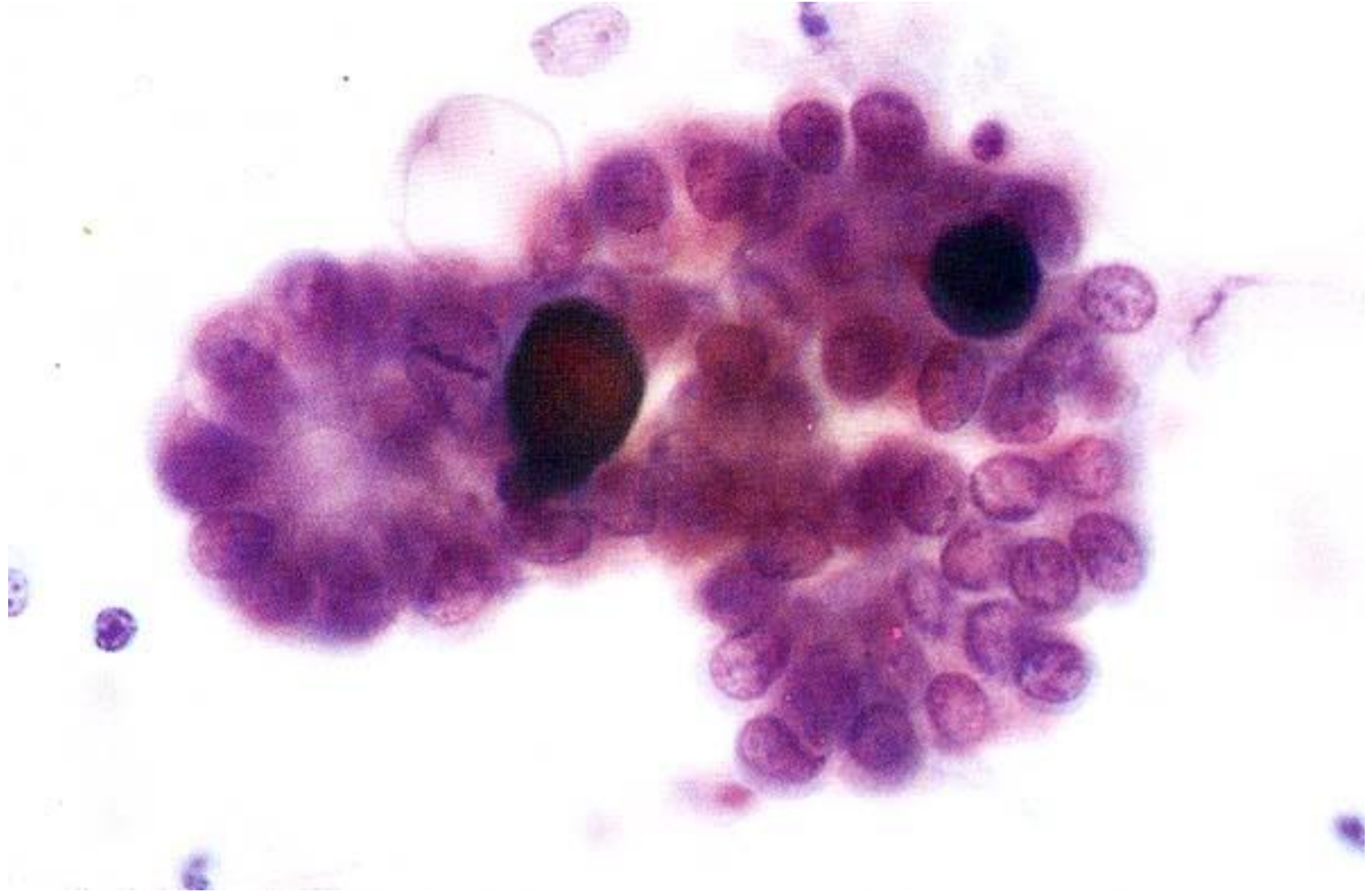
Breast Carcinoma



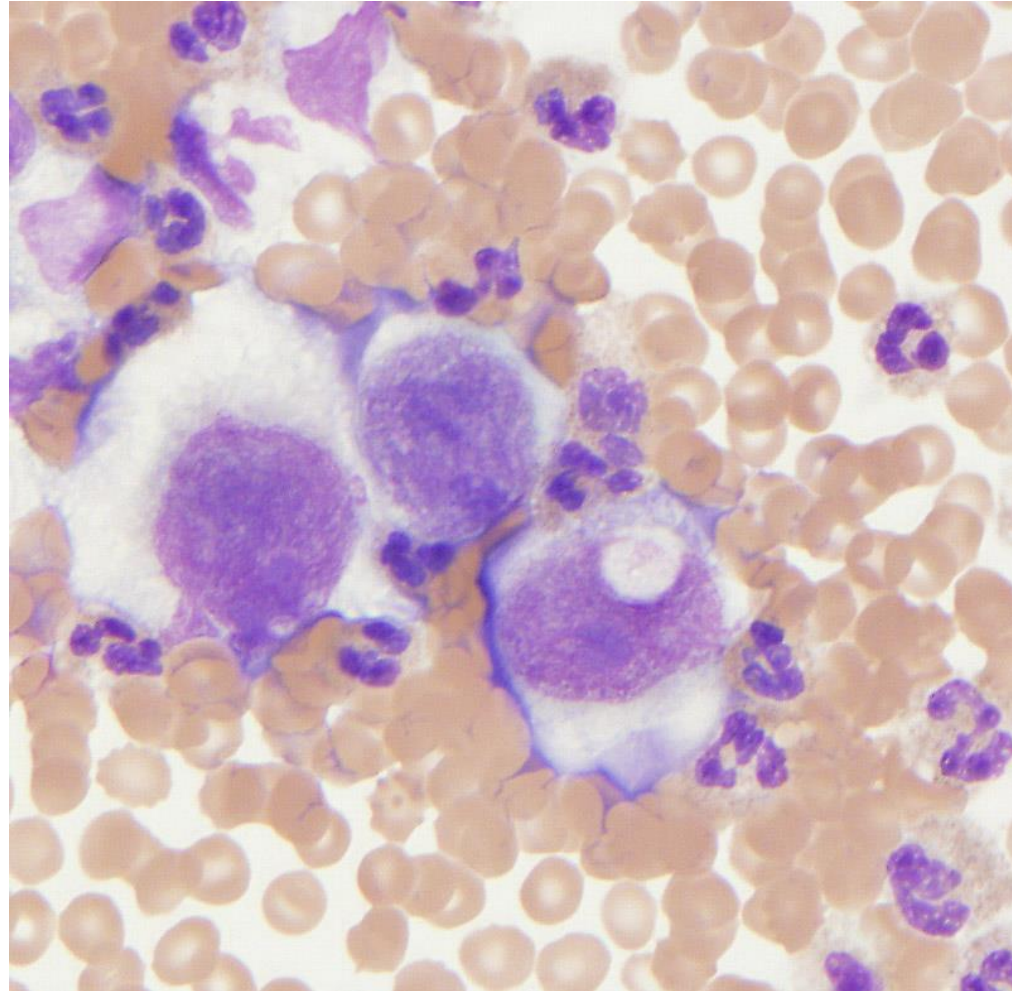
Ovarian Carcinoma



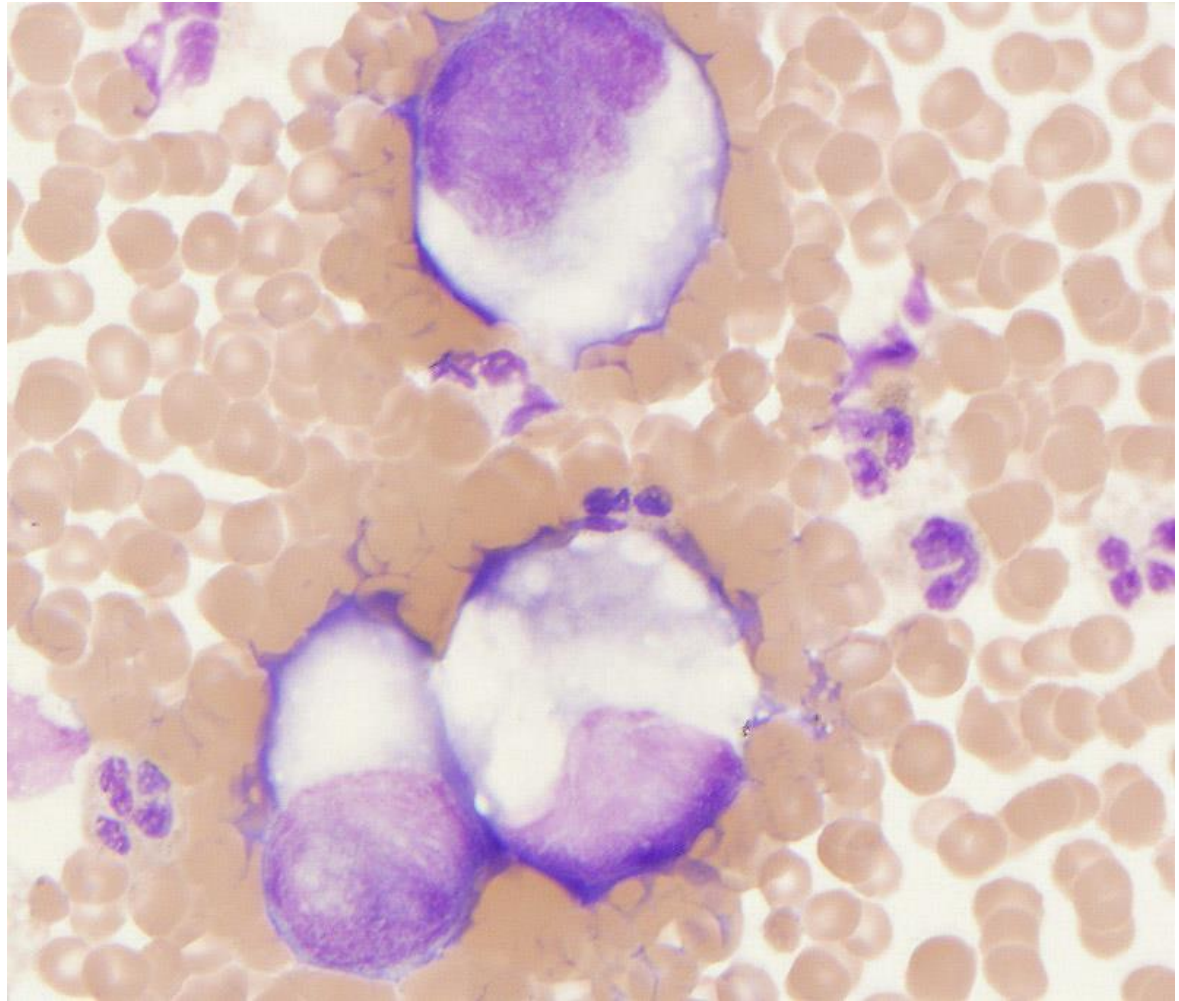
Papillary Serous Carcinoma



Diagnosis?



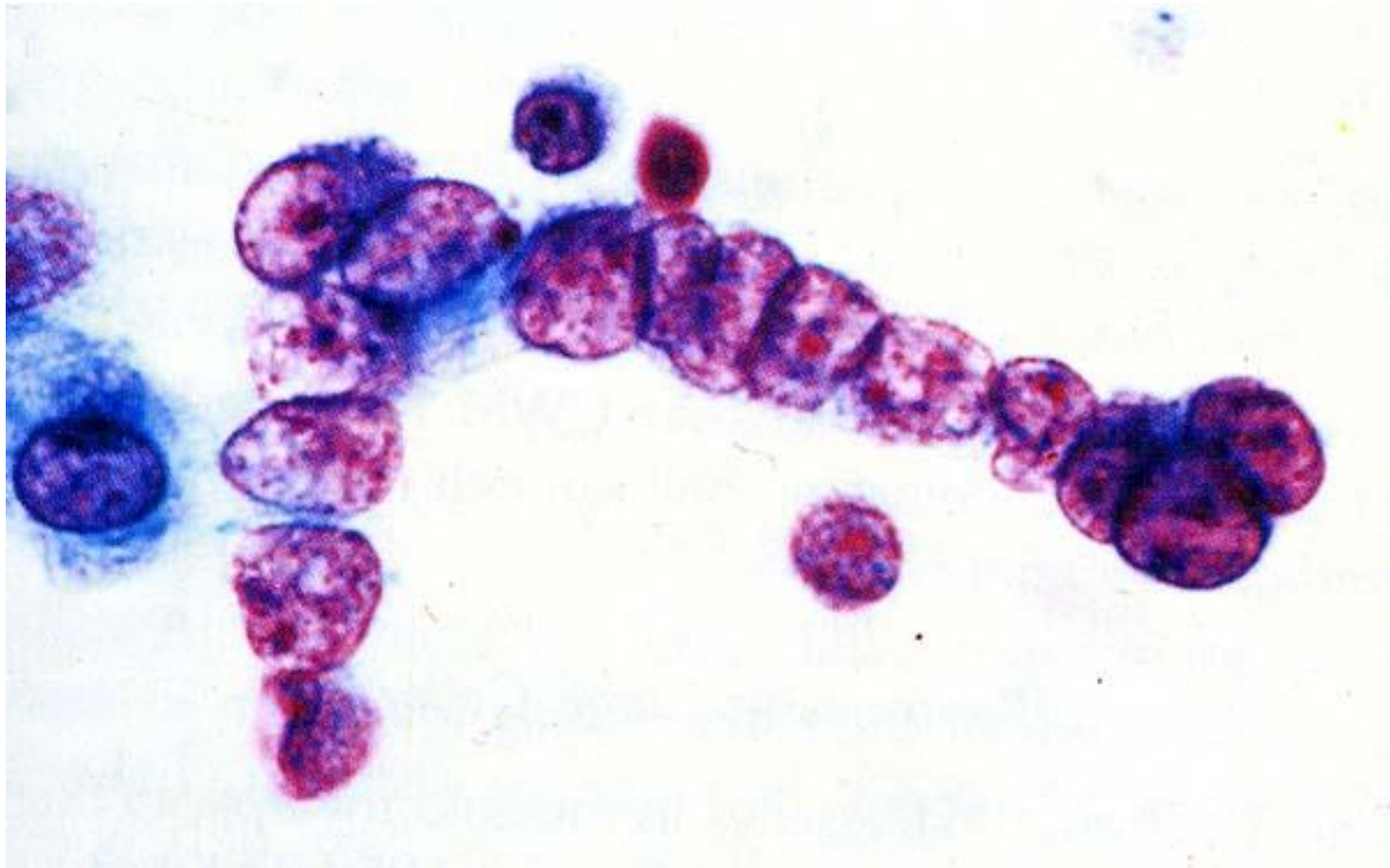
Lung Adenocarcinoma



Diagnosis?



Breast Carcinoma

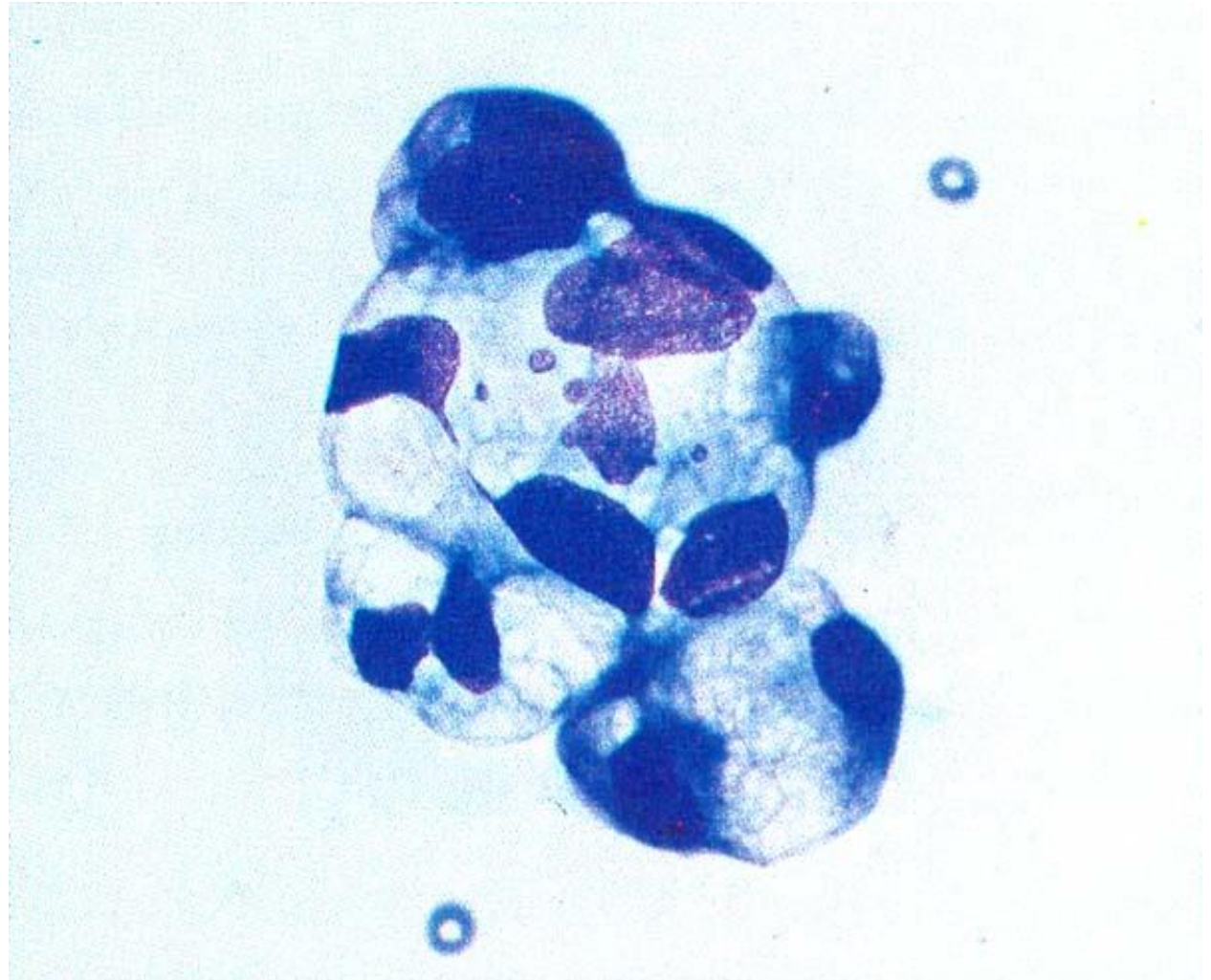


Benign vs. Malignant

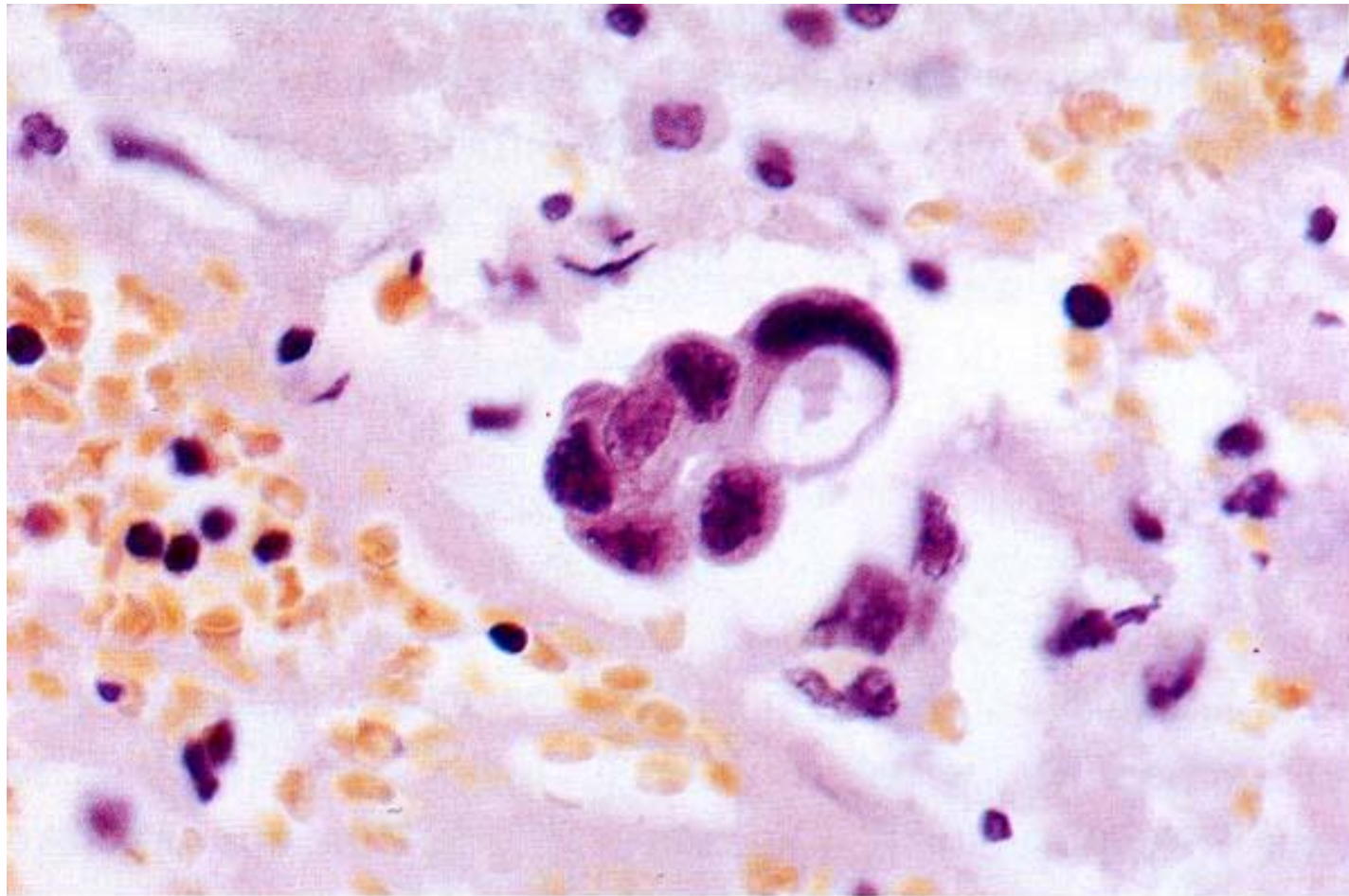
Cytoplasmic vacuoles

- ***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

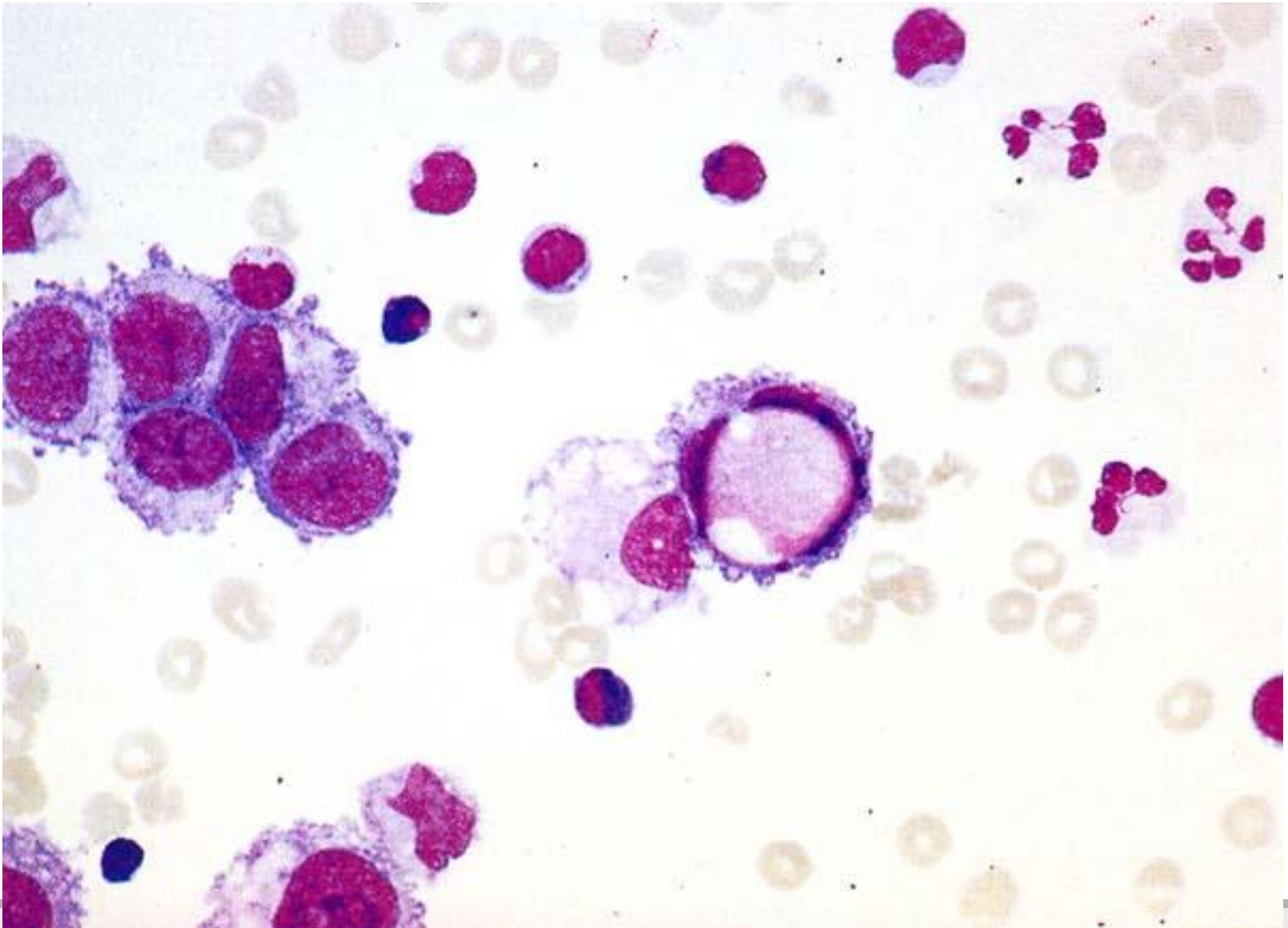
Pancreatic Adenocarcinoma



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



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