Challenges and Lessons Learned – Virtual Microscopy 1

Sarah Wu, MD, PhD

Advances in Cytology and Small Biopsies

June 9th, 2025

Case Vignette #1

- Man in his 70s with a history of prostate cancer, now presenting with a right parotid mass.

- (Show slides)

Differential Diagnosis

- Lymphoma
- Reactive lymph node

What additional testing would you perform?

A. Flow cytometry
B. Cell block and IHC
C. Both A and B
D. None

Flow Cytometry

- Monoclonal B-cells constitute about 85% of total cellularity and possess the following abnormal immunophenotype: kappa-restricted CD19dim+ CD20+ CD5- CD10- CD23- FMC7dim+ CD103- CD11c(dim)+ CD43dim+ sIgM+ sIgG-.

Table 1. Expression Pattern and Frequency of CD5 and CD10 in Common, Mature B-Cell Lymphomas

Immunophenotype	Type of Mature B-Cell Lymphoma	Approximate % Staining	Source, y
$CD5^+/CD10^-$	MCL	93–95	Dorfman and Shahsafaei,[6] 1997; Gualco et al,[7] 2010
	SLL/CLL	80–92	Kurec et al,[4] 1992; Geiler et al,[5] 1991
	LPL	9–43	Hunter et al,[20] 2005; Morice et al,[21] 2009
	MZBCL		
	Splenic MZBCL	20	Gimeno et al,[24] 2005; Matutes et al,[25] 2008
	Nodal MZBCL	8.6	Jaso et al,[22] 2013
	MALT lymphoma	1	Jaso et al,[23] 2012
	De novo DLBCL NOS	10	Tagawa et al,[27] 2005
$CD10^+/CD5^-$	BL	100	Dogan et al,[37] 2000
	FL (low grade)	80	Eshoa et al,[31] 2001
	DLBCL, NOS	10–40	Colomo et al,[46] 2003; Berglund et al,[47] 2005; Visco et al,[48] 2012
	HCL	10–20	Jasionowski et al,[43] 2003; Gupta et al,[44] 2015
	FL (grade 3)	17	Eshoa et al,[31] 2001
	MCL	0–7	Akhter et al,[45] 2015
$CD5^-/CD10^-$	MZBCL		
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	LPL	57–91	Hunter et al,[20] 2005; Morice et al,[21] 2009
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	DLBCL, NOS	50–70	Tagawa et al,[27] 2005; Colomo et al,[46] 2003; Berglund et al,[47] 2005; Visco et al,[48] 2012
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B-cell immunophenotype: CD5- CD10- CD23- FMC7dim+ CD103- CD11c(dim)+ CD43dim+ sIgM+ sIgG-

Abbreviations: BL, Burkitt lymphoma; CLL, chronic lymphocytic leukemia; DLBCL, diffuse large B-cell lymphoma; FL, follicular lymphoma; HCL, hairy cell leukemia; LPL, lymphoplasmacytic lymphoma; MALT, mucosa-associated lymphoid tissue; MCL, mantle cell lymphoma; MZBCL, marginal zone B-cell lymphoma; NOS, not otherwise specified; SLL, small lymphocytic lymphoma.

Wang HY et al. *Arch Pathol Lab Med.* 2017;141:1236–1246.

Summary of Immunohistochemical Profile of Small B-cell Lymphomas

Table 14-7 Classic Immunophenotypic Profile of B-Chronic Lymphoproliferative Neoplasms

Disorder	SIg	CD20	CD22	CD23	CD25	CD5	FMC7	CD11c	CD10	CD79b	CD103	CD200	LEF1	Cyclin D1	SOX11
CLL/SLL	w	w	w	+	–	+	–/w	w	–	–/w	–	+ bright	+	–	–
PLL	+	+	+	–	–	v	v	–	–	+	–	–/+	ND	–	ND
MCL	+	+	+	–, w	–	+	+	–	–	+	–	–	–	+	+
FL	+	+	+	–	–	–	+	–	+	+	–	+ dim/ mod	–	–	–
SMZL	+	+	+	–	–	v	+	v	v	+	–	+ dim/ mod	ND	–	–
HCL	+	+	+	–	+	–	+	+	s	–	+	+ bright	ND	+s	–
LPL	+/CIg	+	+	–	–	–	–	–	–	+	–	+/– dim	–	–	–

References 1, 36, 40, 68, 71, and 199-203.

CIg, cytoplasmic immunoglobulin; *CLL/SLL*, chronic lymphocytic leukemia/small lymphocytic leukemia; *FL*, follicular lymphoma; *HCL*, hairy cell leukemia; *LPL*, lymphoplasmacytic; *MCL*, mantle cell lymphoma; *ND*, not done; *PLL*, prolymphocytic leukemia; *+s*, subset of cases positive; *SMZL*, splenic marginal zone lymphoma, lymphoma; *v*, variable expression; *w*, weakly expressed

B-cell immunophenotype:
CD5- CD10- CD23- FMC7dim+ CD103- CD11c(dim)+ CD43dim+ sIgM+ sIgG-

Chabot-Richards D et al. Chp 14. Hematopathology 2nd ed. 2016.

Cell Block and IHC

- (show slides)

Cell Block Immunophenotype

Positive
- CD20
- PAX5
- Cyclin D1
- SOX11

Negative
- CD5

p53 (wild type pattern)
Ki67 (30-40%)

FISH for *IGH::CCND1*

- IGH::CCND1 rearrangement WAS OBSERVED in 100% of nuclei. In 76% of nuclei, additional copies of IGH, CCND1 and the IGH::CCND1 rearrangement were noted.

What is the diagnosis based on the morphology, flow cytometry, and IHC?

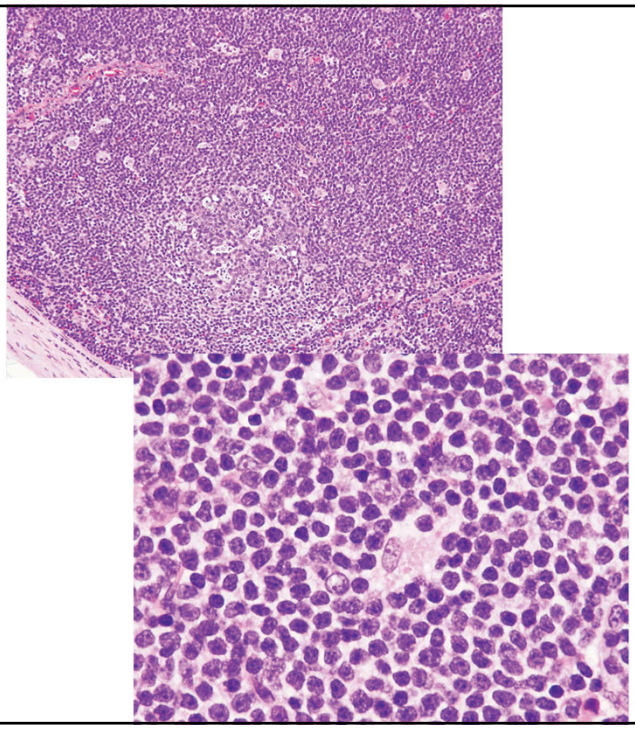
A. Marginal zone lymphoma
B. Mantle cell lymphoma
C. Small lymphocytic lymphoma
D. Follicular lymphoma
E. B-cell lymphoproliferative disorder, NOS

Diagnosis:

Mantle cell lymphoma.

Mantle Cell Lymphoma

- Mature B-cell lymphoma derived from mantle zone of lymph node follicles
- Most arise in lymph nodes, extranodal involvement in spleen, GI, bone marrow, Waldeyer ring
- Older patients (6th-7th decades), M:F 3-4:1
- Small to medium lymphocytes, with hyalinized small vessels and epithelioid histiocytes in the background
- IHC
  - +: B cell markers (CD19, CD20, PAX5), CD5, cyclin D1, SOX11, BCL2, CD43
  - -: CD10, BCL6, CD23
- *CCND1::IGH* translocation: t(11;14)(q13;q32) in most cases
  - Rare cyclin D1-negative cases have alterations involving *CCND2* or *CCND3*, SOX11 will be positive
- Prognostic markers
  - Ki67 proliferation rate: >30% is worse
  - *TP53* mutation – NGS can be performed



WHO 5th edition, Mantle cell lymphoma

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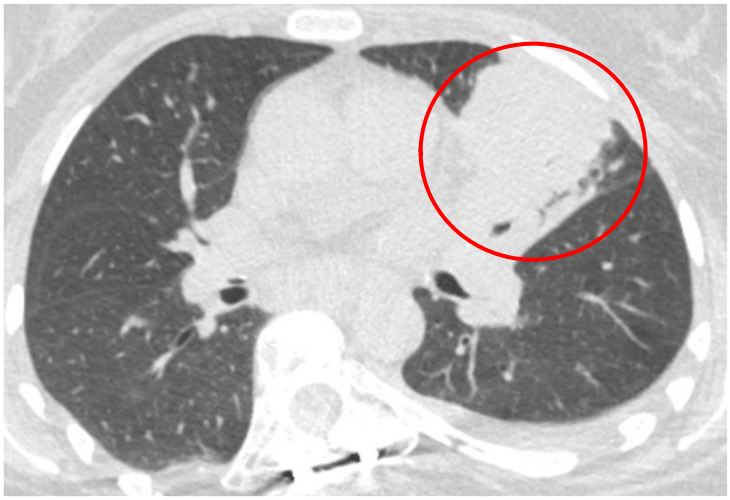
Take Home Points

- CD5 negative mantle cell lymphoma is uncommon but a potential diagnostic pitfall
  - Mantle cell lymphoma is important to avoid missing since it has a worse prognosis than marginal zone lymphoma
- Utilization of cyclin D1 IHC can be helpful in the routine workup of small B-cell lymphoma cytology cases.
- Recommended small B-cell IHC panel:
  - CD3, CD20 or PAX5, CD5, CD10, cyclin D1, Ki67
    - SOX11 can be performed if cyclin D1 is positive
  - For core biopsy specimens, BCL2, BCL6 for germinal centers, CD21/CD23 for follicular dendritic cell meshworks

Case Vignette #2

- Woman in 50s presenting with rapidly increasing lymphocytosis (over 250 K/uL), cough
- Frequently travels to Arizona to visit family
- Never smoker

CT Chest



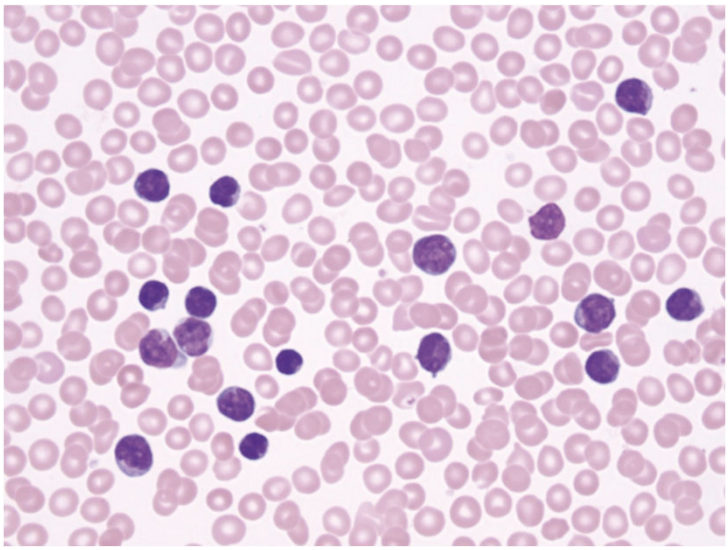
- 5 cm mass-like consolidation in LUL
- Multiple enlarged mediastinal and left hilar lymph nodes
- Splenomegaly

Differential Diagnosis

- Infection
- Lung primary malignancy
- Lymphoma
- Leukemia

Flow Cytometry of Peripheral Blood

- Atypical T cells (97% of gated events; 83% of total events) that are positive for CD3, CD4 (dim), CD2(slightly dim), CD5, and CD7(variable), and negative for CD8, CD16, and CD56. The peripheral blood smear shows marked lymphocytosis consisting of medium-sized lymphocytes with round to mildly irregular nuclei, moderately condensed chromatin, variably visible nucleoli and moderate amounts of basophilic cytoplasm, with frequent cytoplasmic blebs present.



WHO 5th edition, T-prolymphocytic leukemia

TCR Clonality Studies on Peripheral Blood

- RESULT:
  Clonal pattern.
  A clonal T cell receptor gamma chain gene rearrangement is identified (192 and 195 bp)

Cytogenetics on Peripheral Blood

- 45~47,XX,-5,-6,+8[4],add(13)(q34),t(14;14)(q11;q32.1),-17,+r,+2mar,inc[cp7]/ 46,XX[2].nuc ish(D8Z2,MYC)x3[46/100],(TRA/Dx2)(5'TRA/D sep 3'TRA/Dx1)[98/100], (TP53x2-3,D17Z1x1)[36/100]

- Seven of 9 metaphases from this unstimulated peripheral blood specimen contained a complex clone, including trisomy 8, a translocation 14;14, and monosomy 17, among other aberrations.
- FISH confirmed that the t(14;14) results in T-cell receptor alpha/delta locus rearrangement, which is a common finding in a subset of T-cell leukemias and lymphomas. While this rearrangement is suggestive of TRA/D::TCL1, this analysis did NOT evaluate the presence/absence of TCL1 rearrangement (as this FISH probe is not available in this laboratory), and thus the specific rearrangement partner the T-cell alpha/delta locus is uncertain.

EBUS of LUL Performed

- (Show slides)

Other Testing

- Positive fungal cultures of LUL mass
- Positive serology for Coccidioides (IgM and IgG)

Diagnosis:

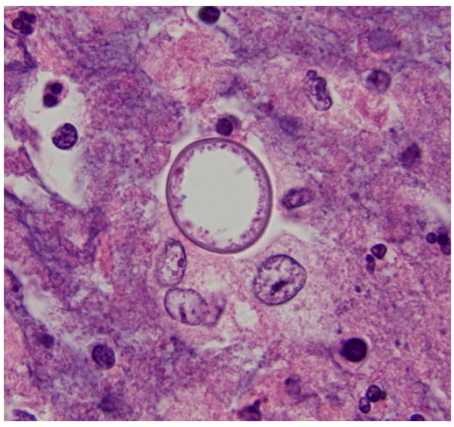
Granulomatous inflammation with focal necrosis and few spherules with endospores, consistent with *Coccidioides* species.

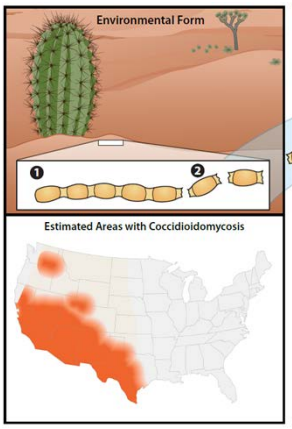
Background of lymphocytes consistent with involvement by the patient's known CD4+ T cell lymphoproliferative disorder, most consistent with T-prolymphocytic leukemia (T-PLL).

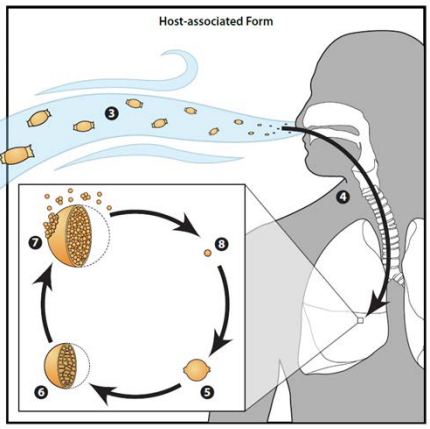
Follow up

- Patient recalled possible exposure to construction dust from houses around where she was staying in Arizona
- Treated with fluconazole
- Treated with Campath for T-PLL, with plan for eventual allogeneic stem cell transplant

Coccidioidomycosis





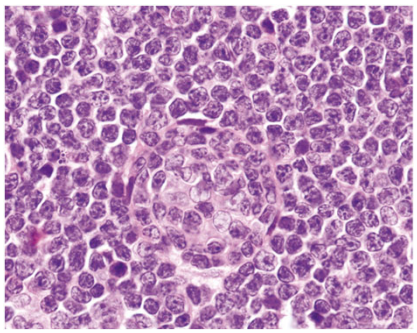


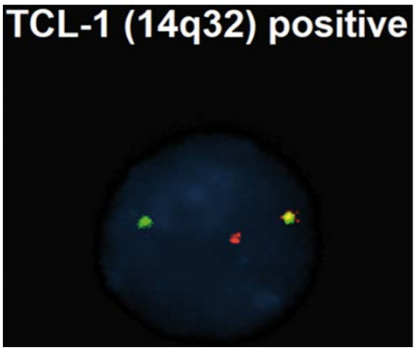
- Dimorphic fungus grows as mycelium in soil and spherule in host
- Endemic to southwest and western US
- Spherules with endospores (broken ping-pong ball)
- Background granulomatous inflammation

https://www.cdc.gov/fungal/diseases/coccidioidomycosis/causes.html

T-prolymphocytic leukemia

- Rare T-cell leukemia characterized by *TCL1A* rearrangements to TR gene enhancer loci of TRA/TRD, t(14;14)
  - Complex karyotypes seems in 70-80% of cases.
- Clinical presentation of marked leukocytosis (>100 K/uL), can see hepatosplenomegaly and lymphadenopathy
- Immunophenotype commonly CD4+, co-expression of CD4 and CD8 can be seen and is characteristic
- TCL1 IHC is helpful for diagnosis
- DDX: T-cell lymphoma with leukemic progression (ATLL, Sezary, T-LGL leukemia)





WHO 5th edition, T-prolymphocytic leukemia

Thank you for participating!

Questions?

Challenges and Lessons Learned

Melanie C. Kwan, MD
Fellow in Cytopathology
Mass General Brigham

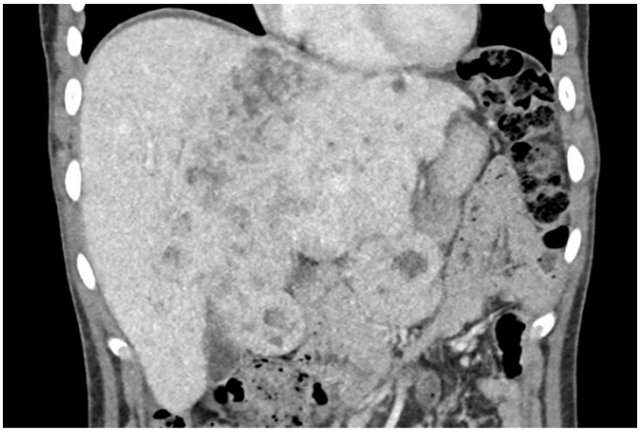
Case Presentation Outline

- Clinical history
- Imaging findings
- Cytology findings
- Differential
- Follow-up

Clinical history

- 30s F
- Presented with 6 months abdominal pain

CT abdomen/pelvis

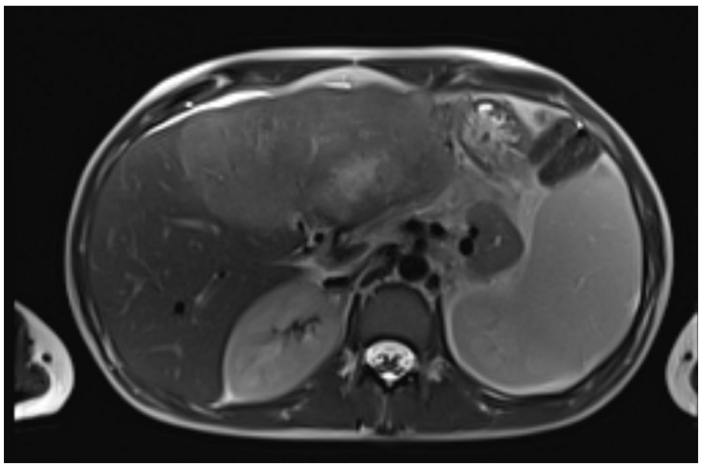


EGD/EUS

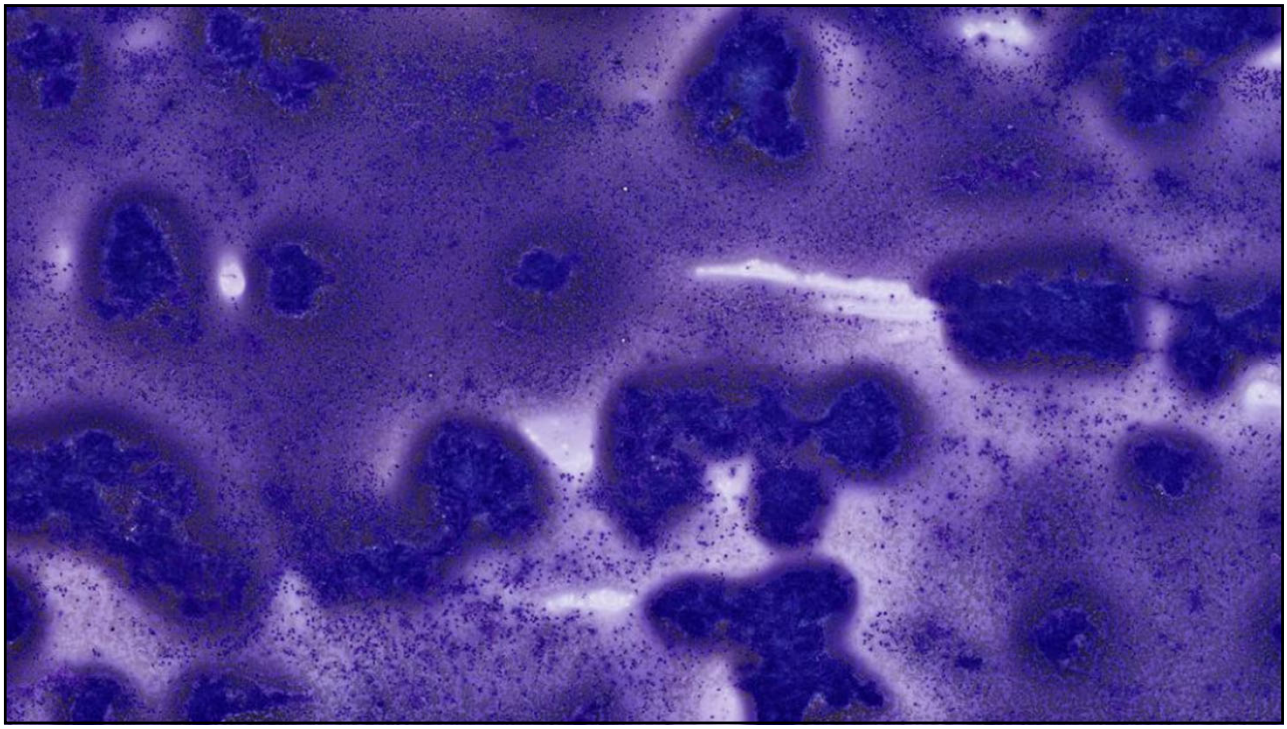
- Oval mass in pancreatic body
- Hypoechoic
- 63.5 mm by 40.0 mm
- Well-defined borders
- No pancreatic ductal dilation
- FNA performed using a transgastric approach

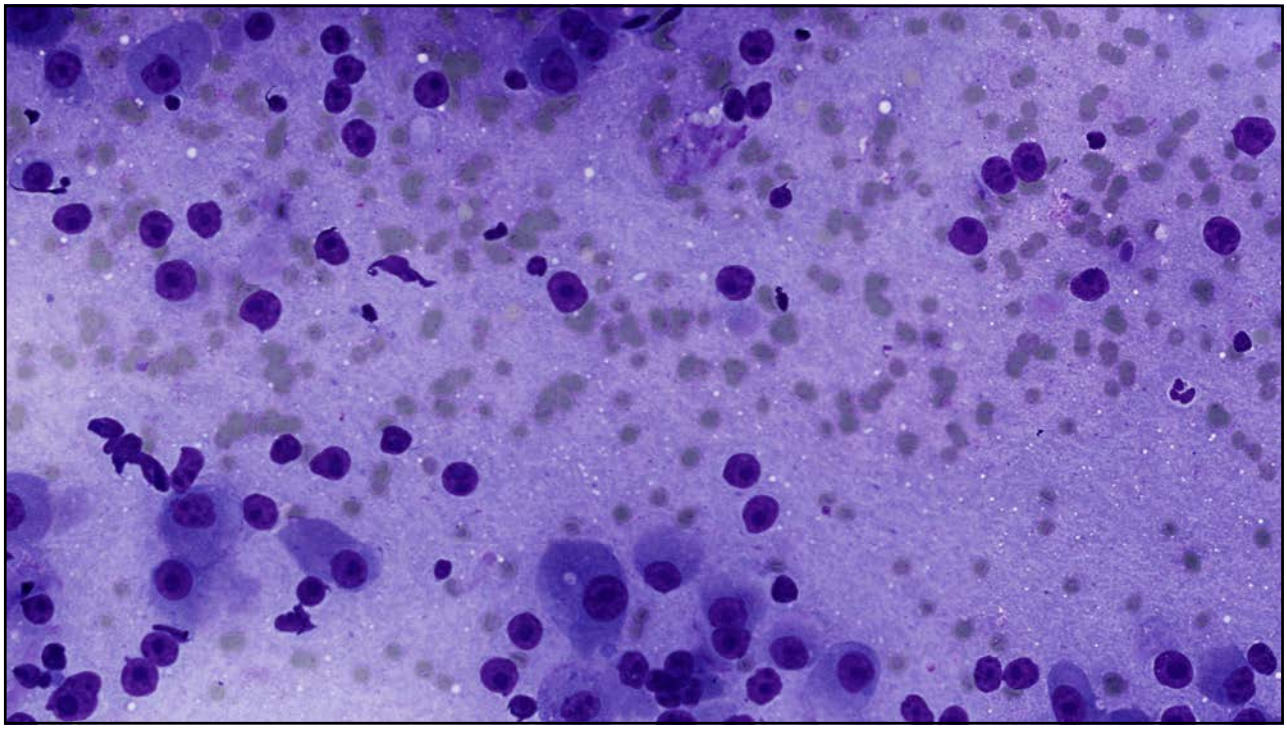
- Abnormal areas in left lobe of the liver
- Hyperechoic
- Representative area 41 mm
- Poorly defined border
- FNA performed using a transgastric approach

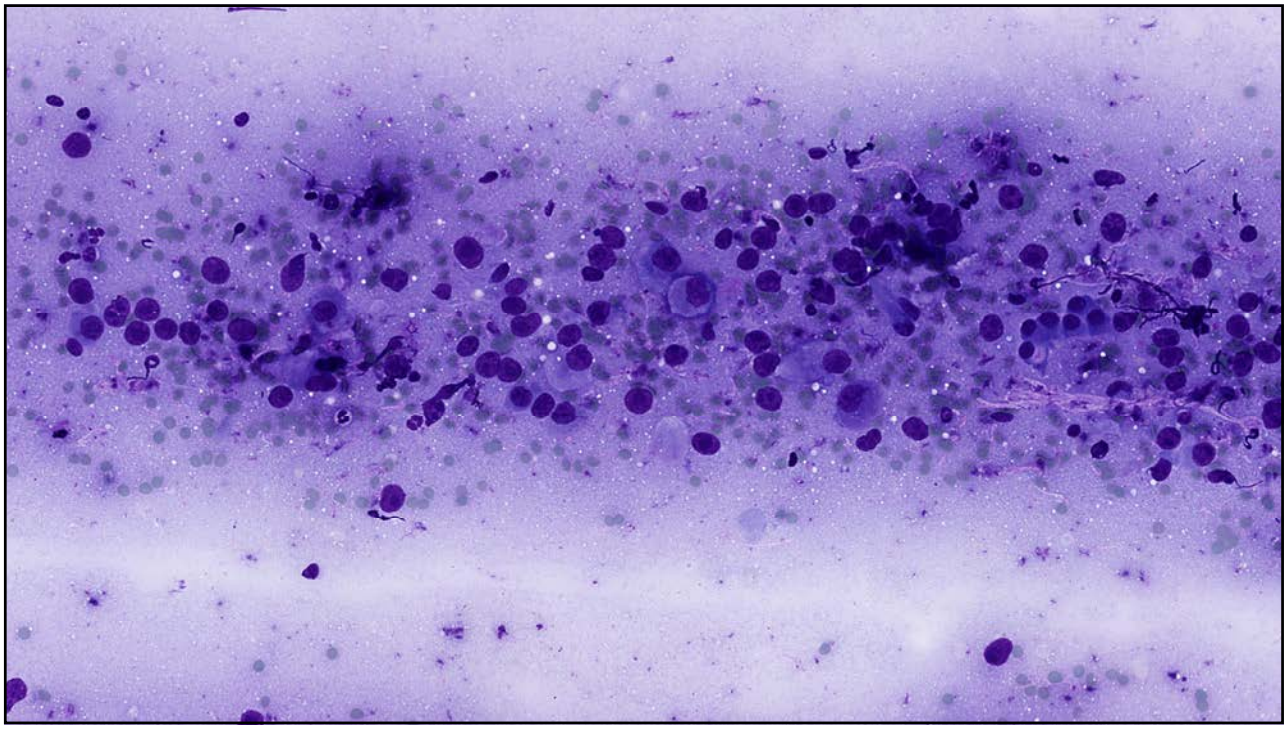
Imaging - MR

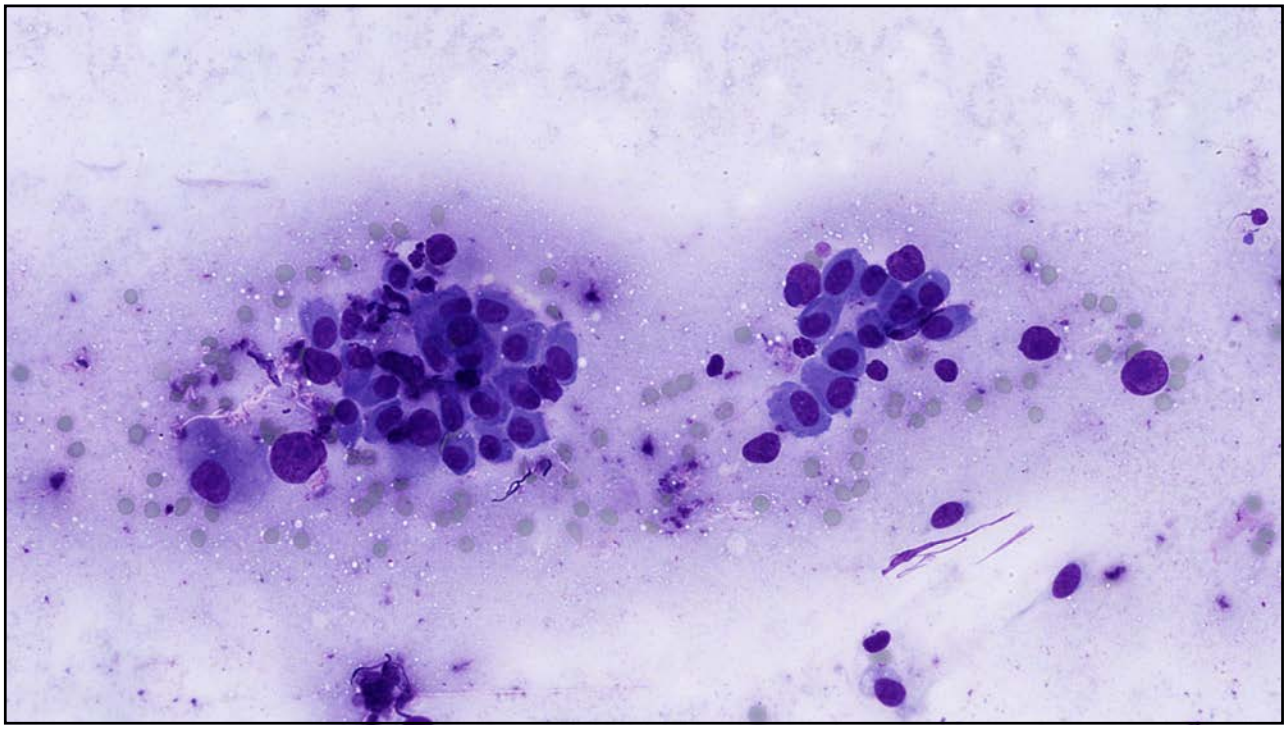


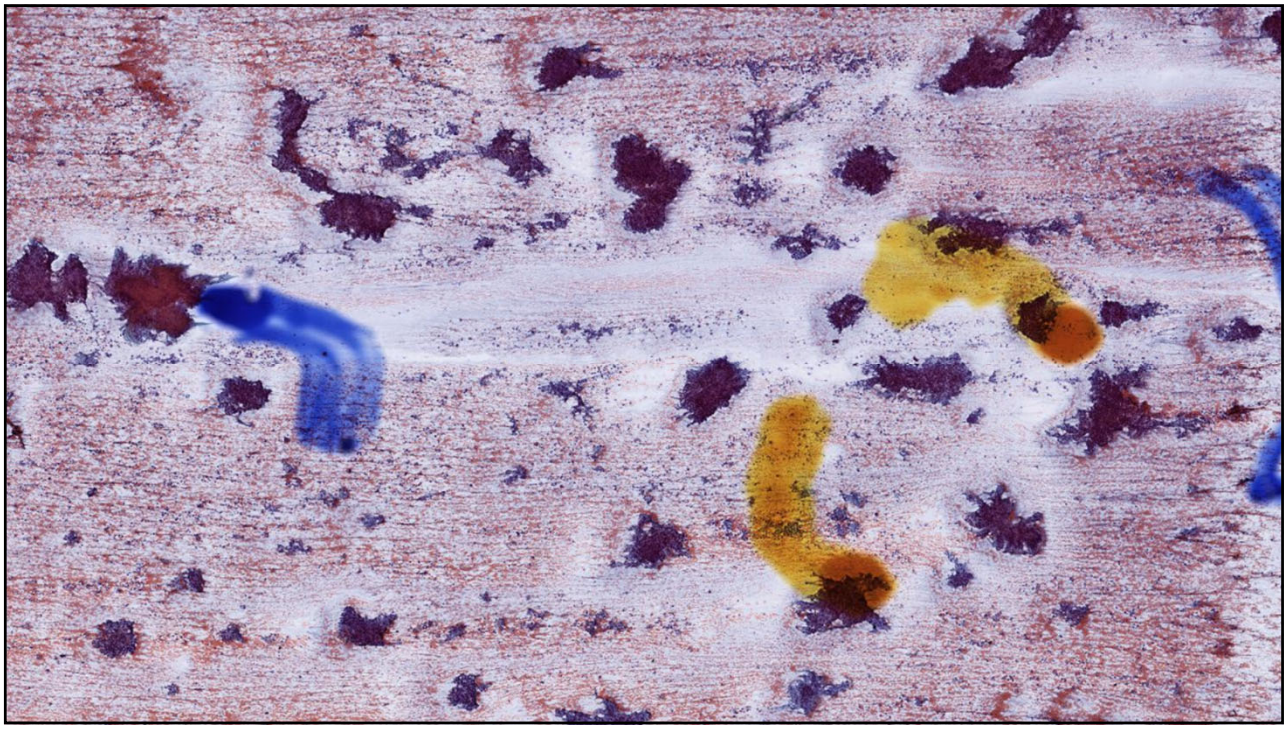
Pancreas FNA

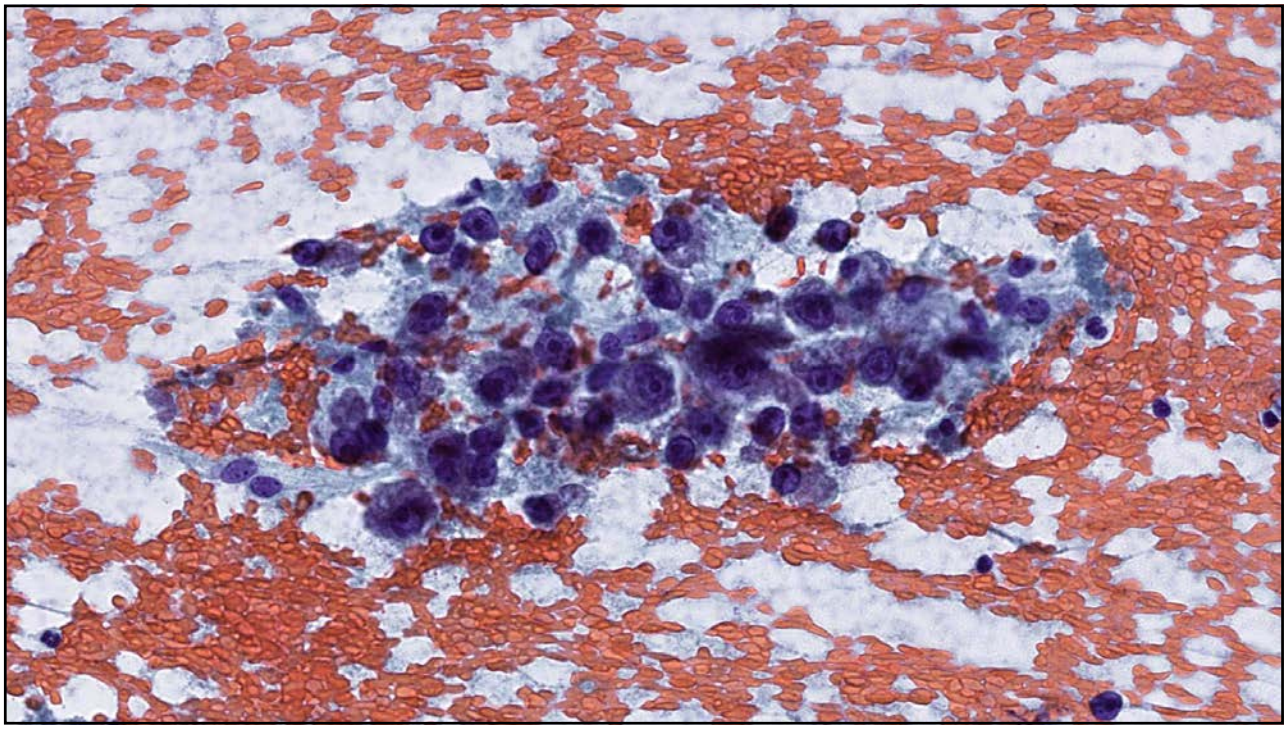


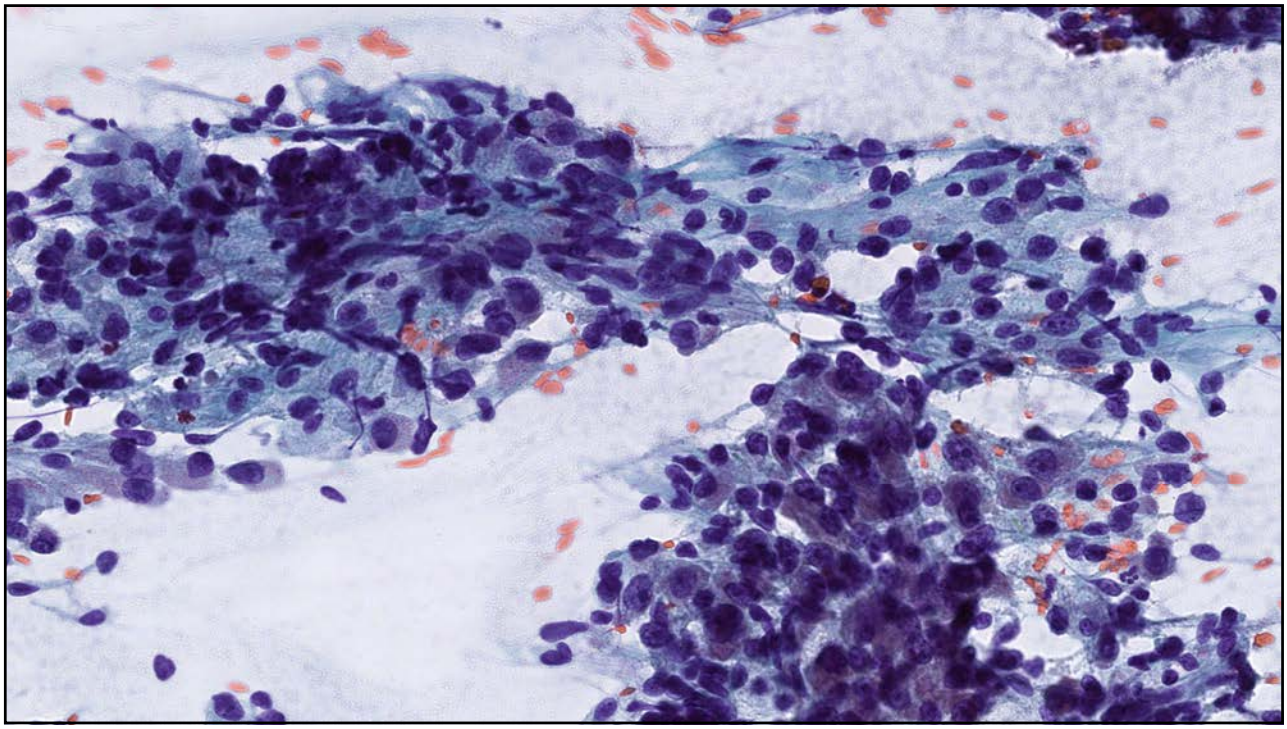


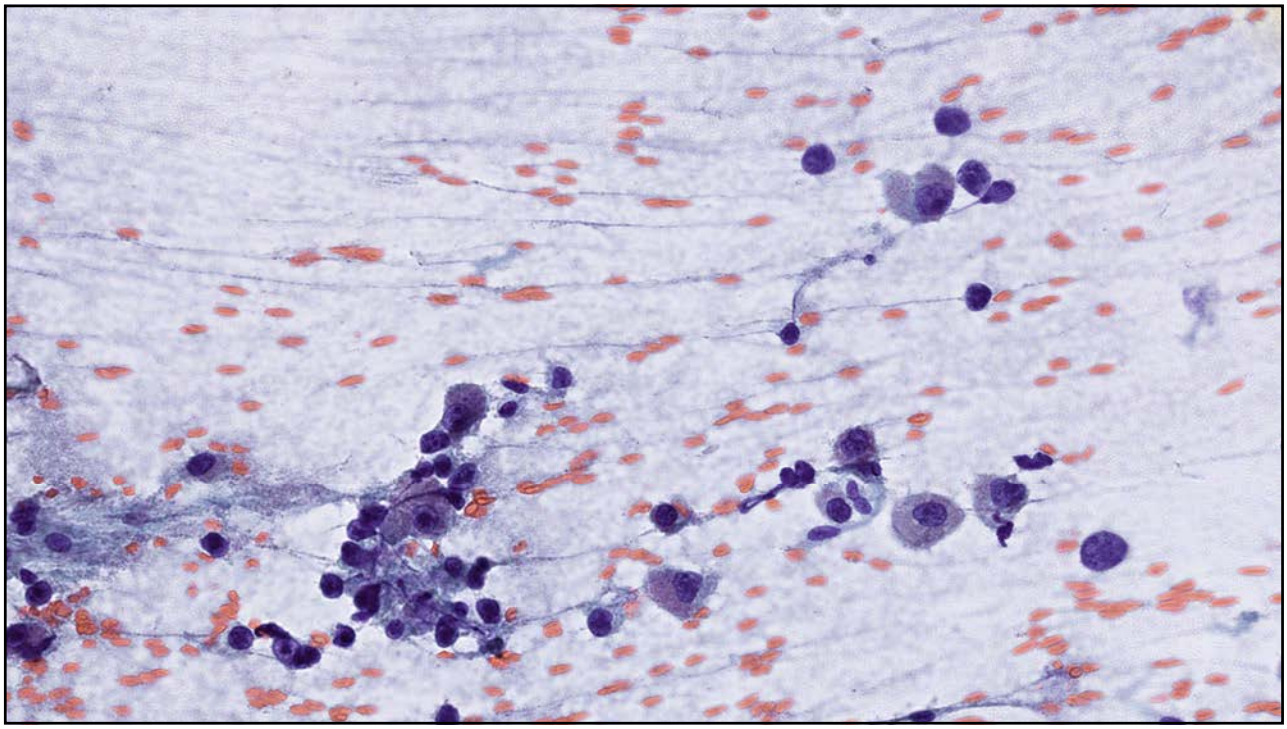






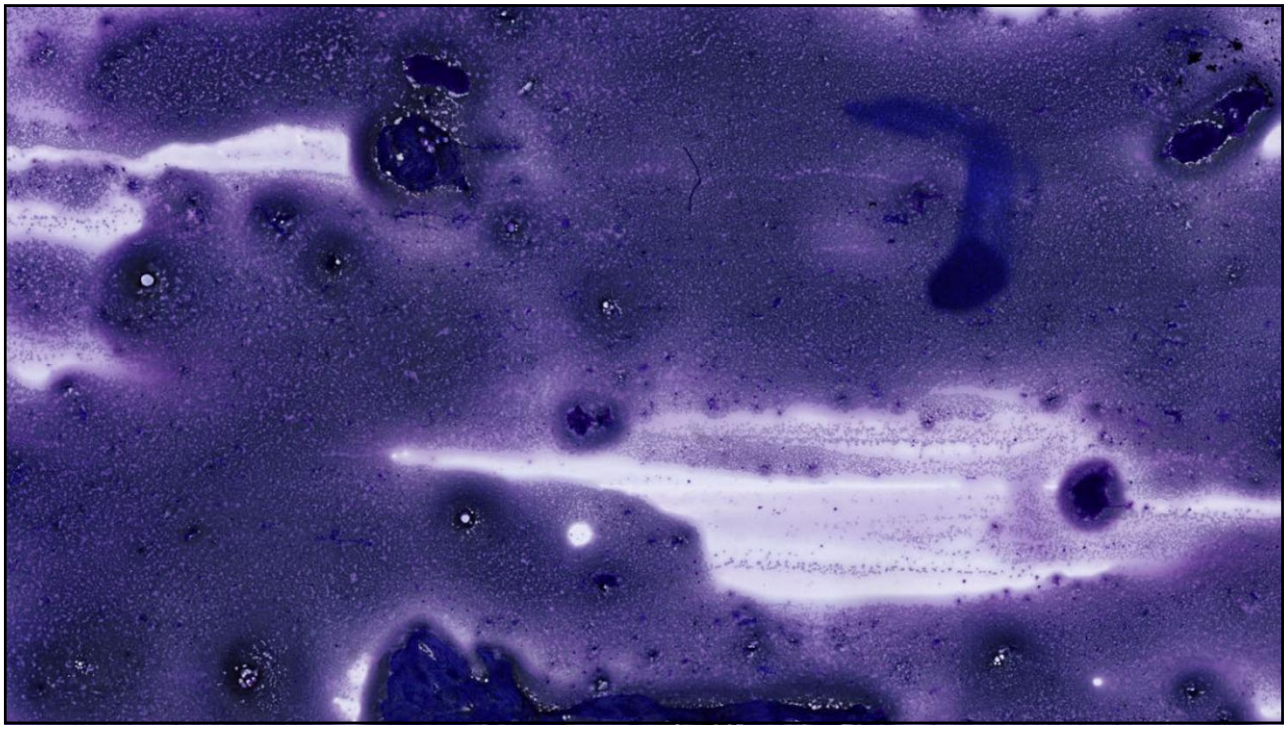


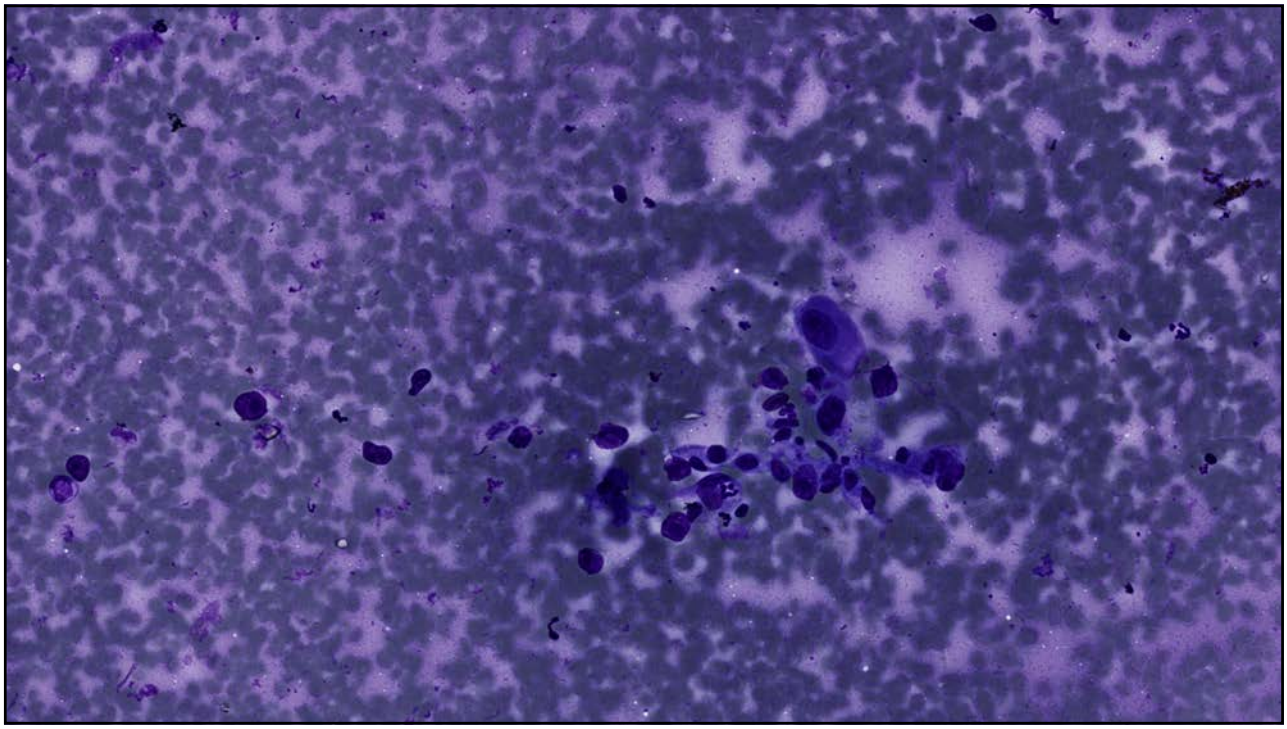


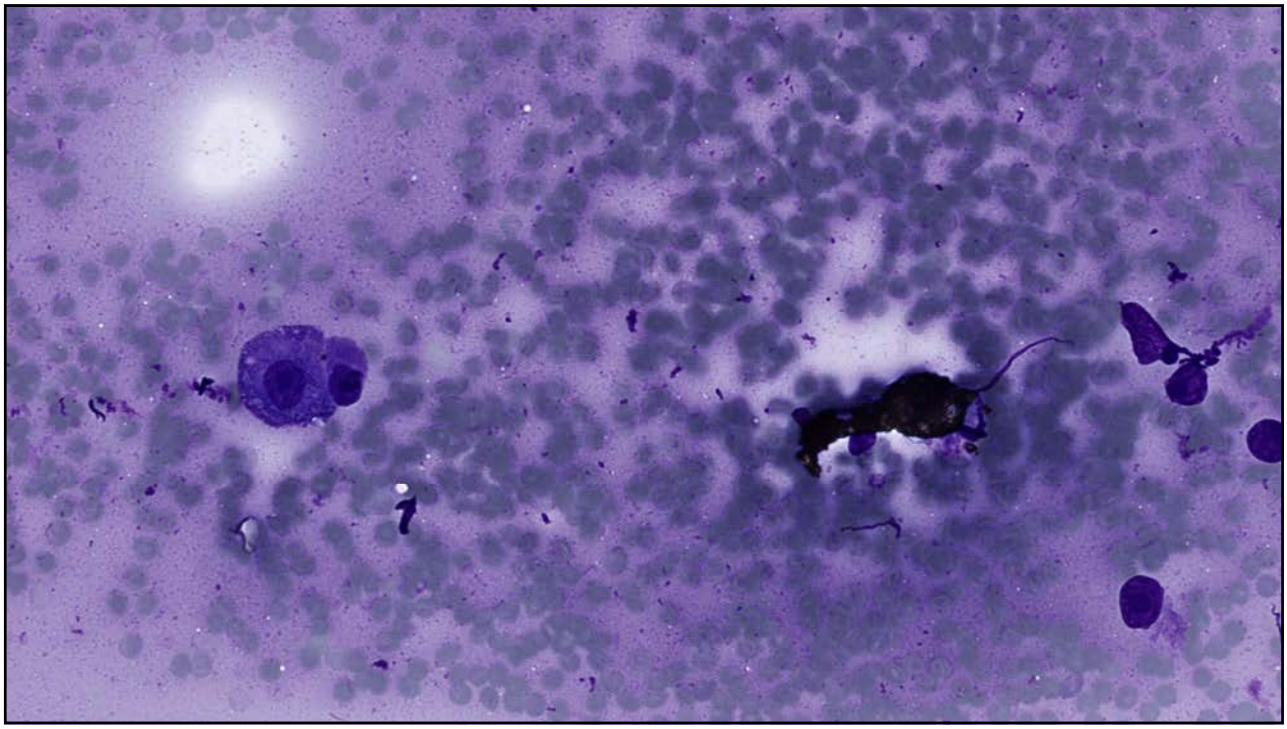


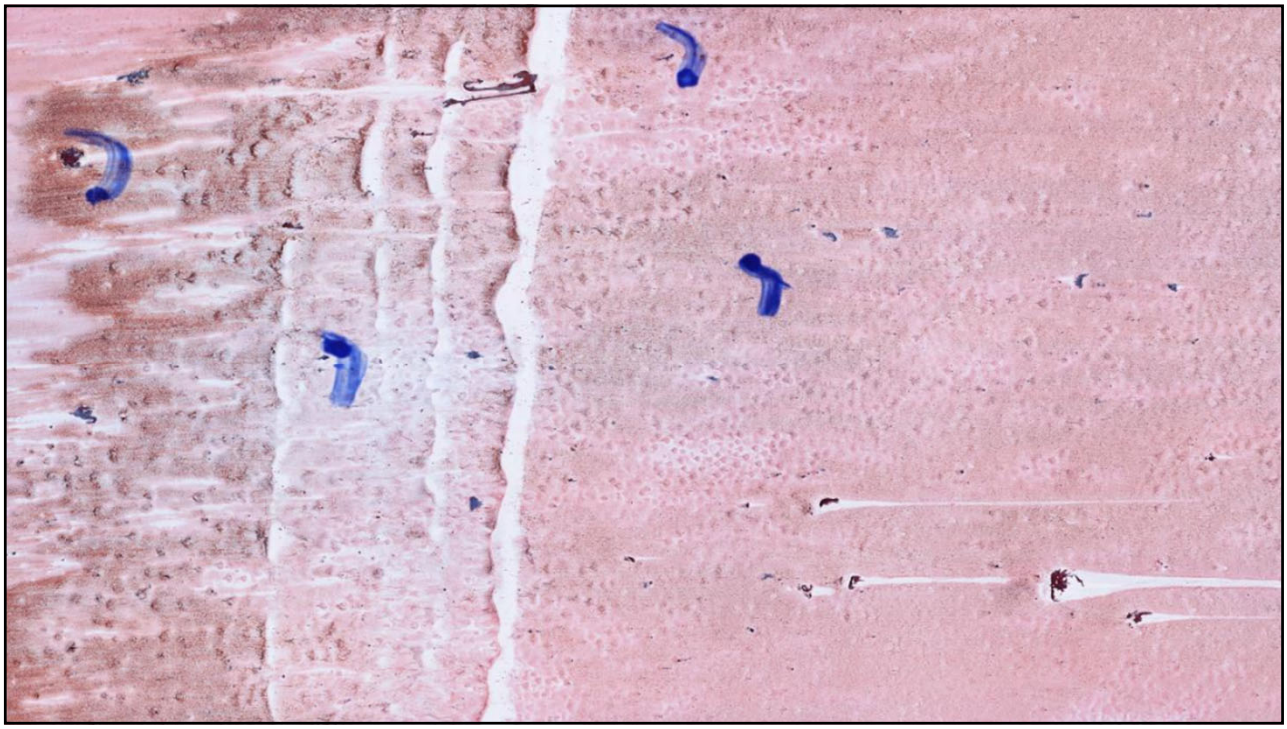
Summary of cytologic findings

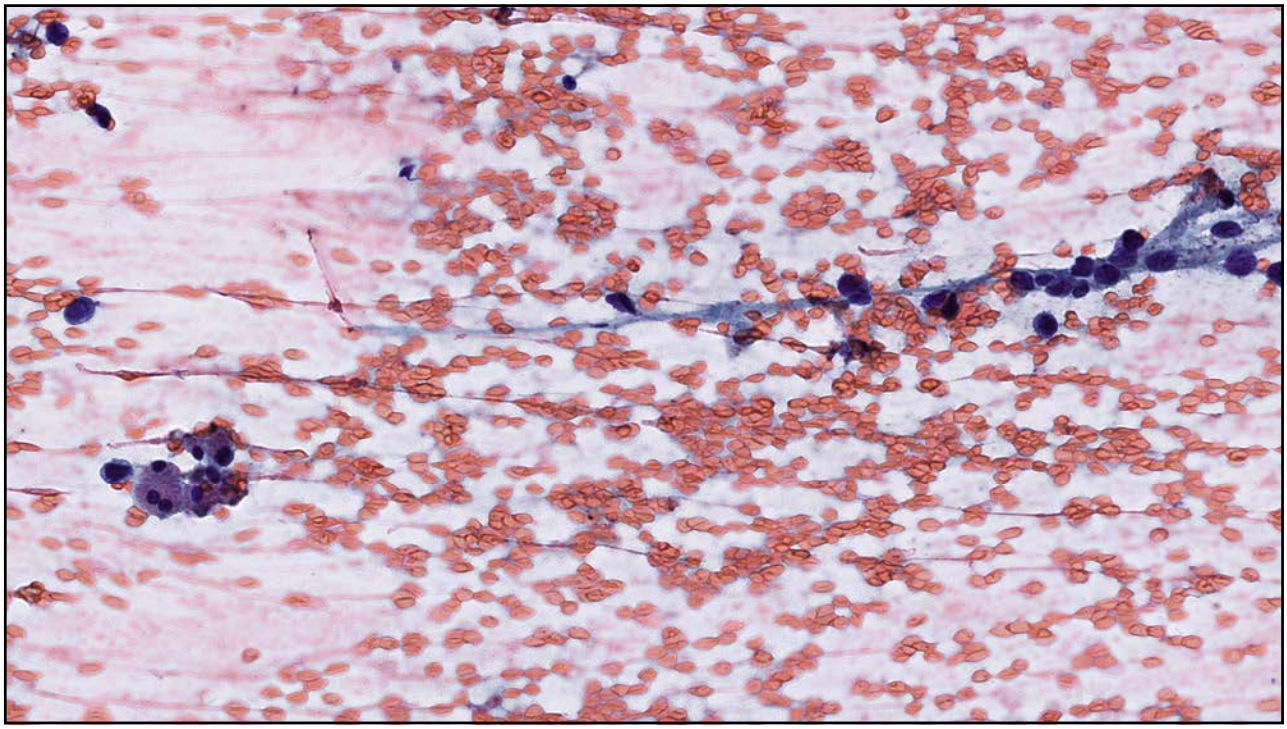
Liver FNA

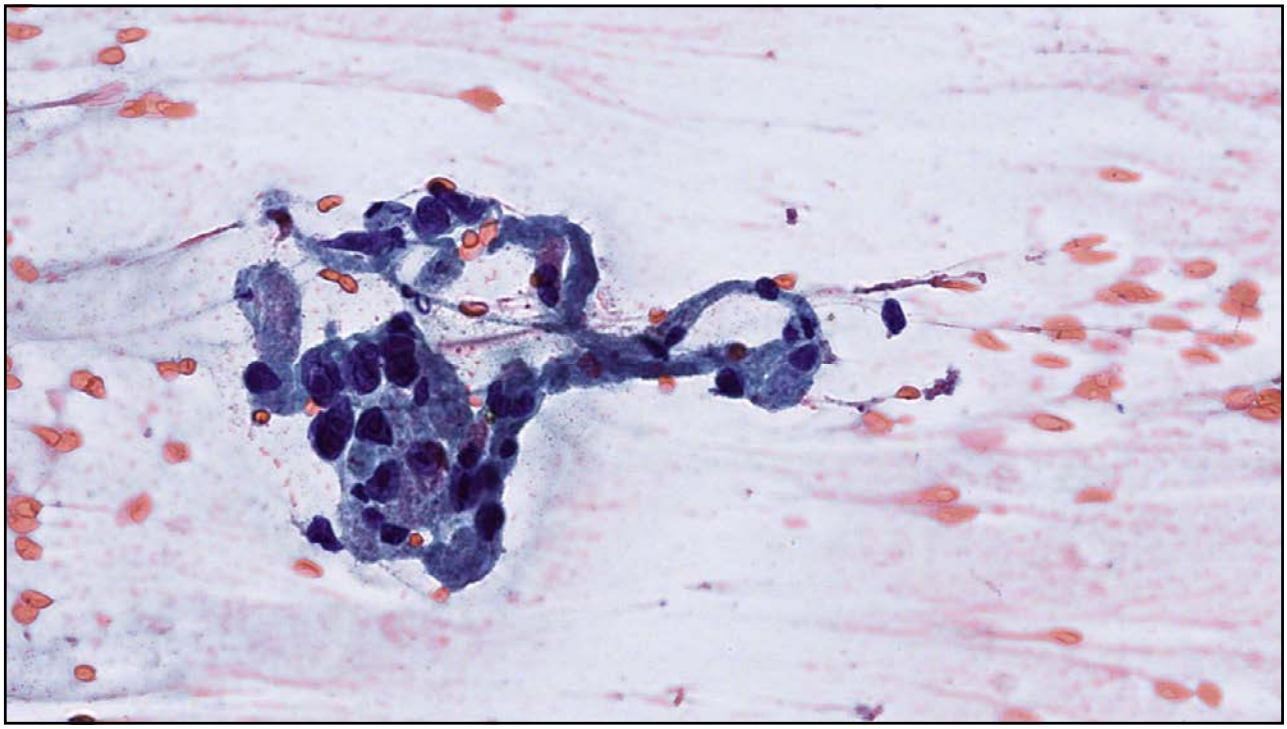


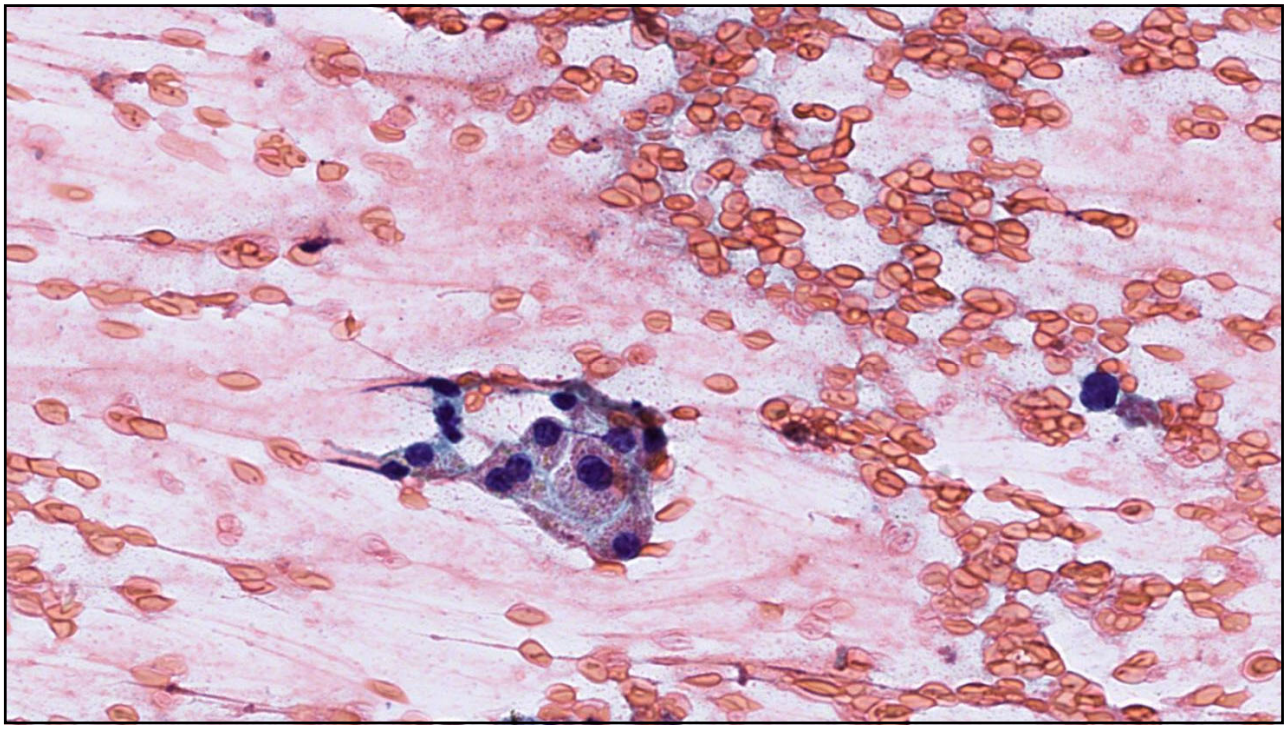












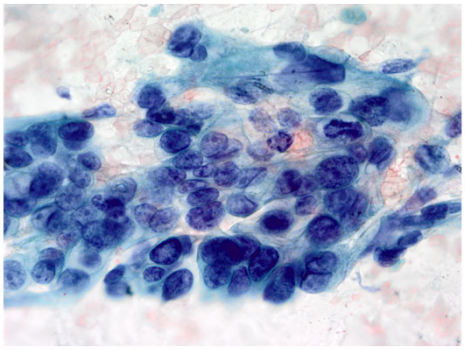
Summary of cytologic findings

Pancreatic Lesions

- Solid
  - Pancreatic Ductal Adenocarcinoma (PDAC)
  - Pancreatic Neuroendocrine Neoplasm
    - Neuroendocrine Tumor (PanNET)
    - Neuroendocrine Carcinoma (PanNEC)
  - Acinar Cell Carcinoma
  - Solid Pseudopapillary Neoplasm (SPN)
  - Metastases
  - Ectopic spleen
- Cystic
  - Neoplastic Mucinous Cysts
    - Intraductal Papillary Mucinous Neoplasm (IPMN)
    - Mucinous Cystic Neoplasm (MCN)
  - Non-neoplastic Mucinous Cysts
    - Serous Cystadenoma (SCA)
    - Pseudocyst
    - Lymphoepithelial Cyst (LEC)

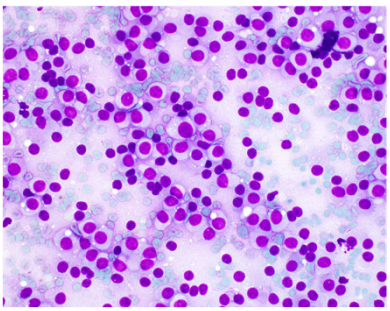
Pancreatic Ductal Adenocarcinoma

- Highly cellular smears with tissue fragments
- Loosely cohesive or crowded tissue fragments
- Irregular nuclear spacing and loss of polarity
- Mucinous cytoplasm
- Hypochromatic nuclei



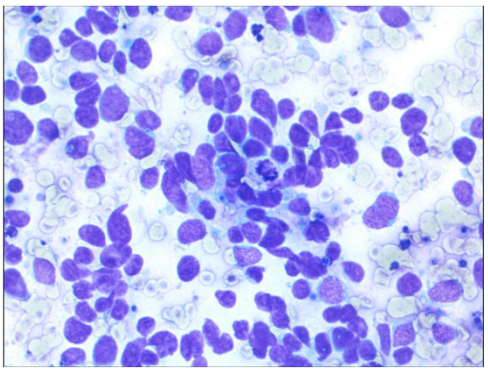
Pancreatic Neuroendocrine Tumor

- Highly cellular smears
- Loosely cohesive and occasionally 3D tissue fragments and dispersed individual cells
- Epithelioid or plasmacytoid cells, occasionally with minimal cytoplasm
- Round to ovoid nuclei
- Smooth, dense nuclear membranes, coarsely stippled salt-and-pepper chromatin
- Dense cytoplasm, clean background



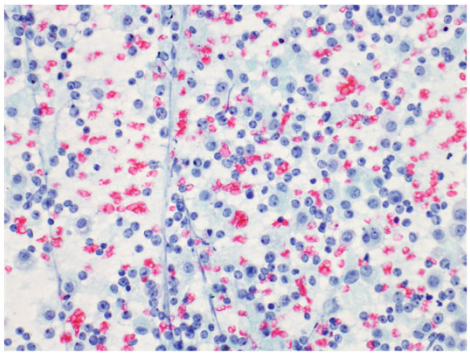
Pancreatic Neuroendocrine Carcinoma

- Highly cellular smears
- Loose crowded tissue fragments, occasionally 3D and/or single cells
- Necrosis
- Angulated nuclei with molding



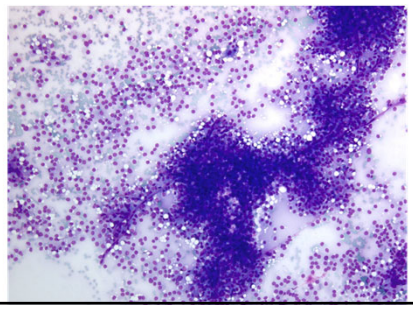
Acinar Cell Carcinoma

- Hypercellular smears with dense 3D tissue fragments, single cells
- Lobulated, trabecular, and small acinar architecture
- Large nuclei, prominent nucleoli
- Granular cytoplasm
- Granular background



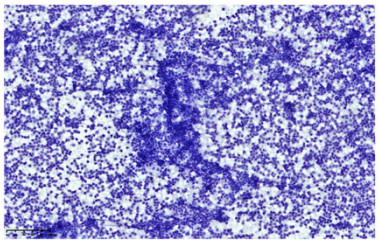
Solid Pseudopapillary Neoplasm

- Highly cellular smears with branching papillary fronds and single cells
- Three layers in papillary fronds
- Round to oval nuclei with smooth nuclear membranes
- Finely granular chromatin
- Prominent nucleoli
- Background cyst debris



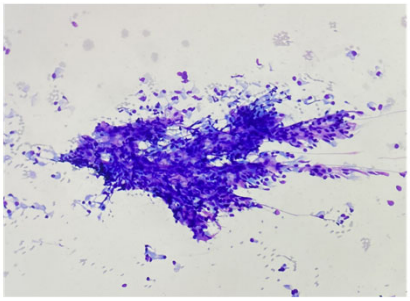
Ectopic Spleen

- Well-circumscribed, vascular nodule in pancreatic tail
- Mimics PanNET on imaging
- Numerous lymphocytes and lymphoid aggregates
- CD8 highlights splenic littoral cells lining vascular spaces in red pulp
- Can be mistaken for a low-grade lymphoma



Metastasis

- Uncommon
- 2% to 5% of pancreatic malignancies
- Most common primary sites: kidney, lung, breast, colon
- Renal cell carcinoma most common
- Can mimic a primary pancreatic lesion

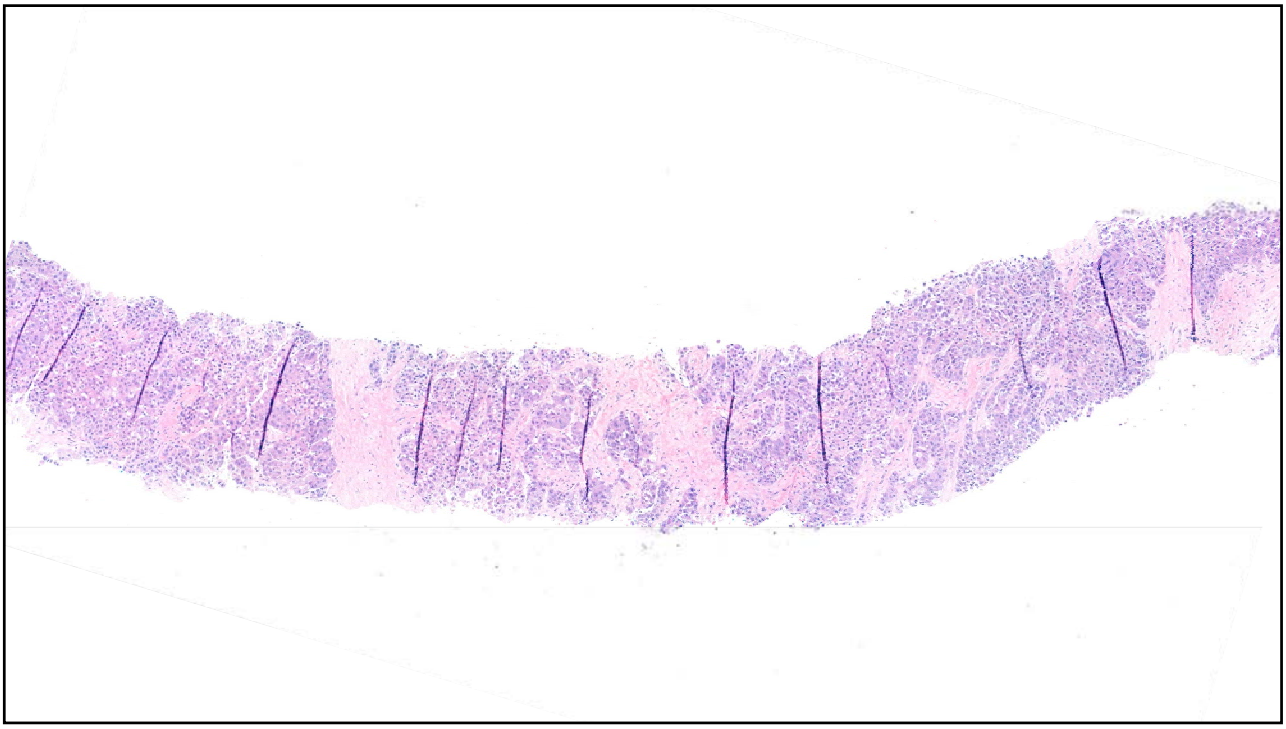


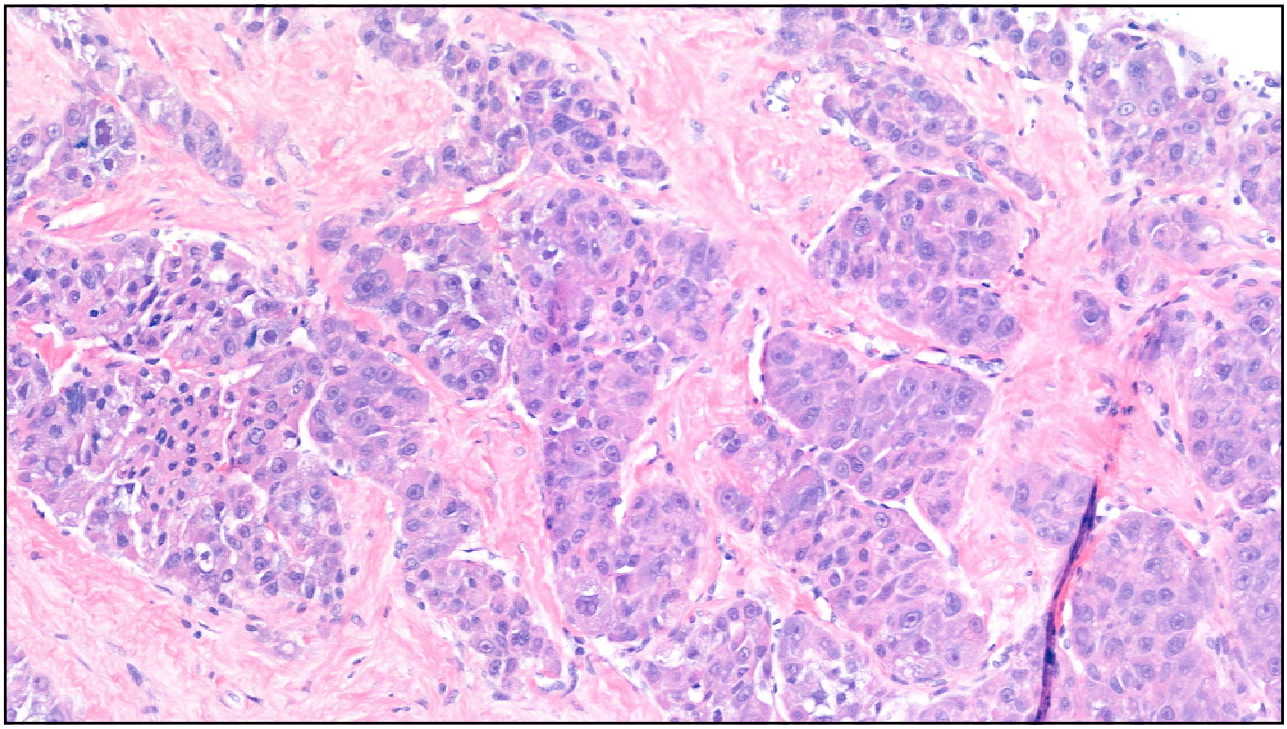
Differential

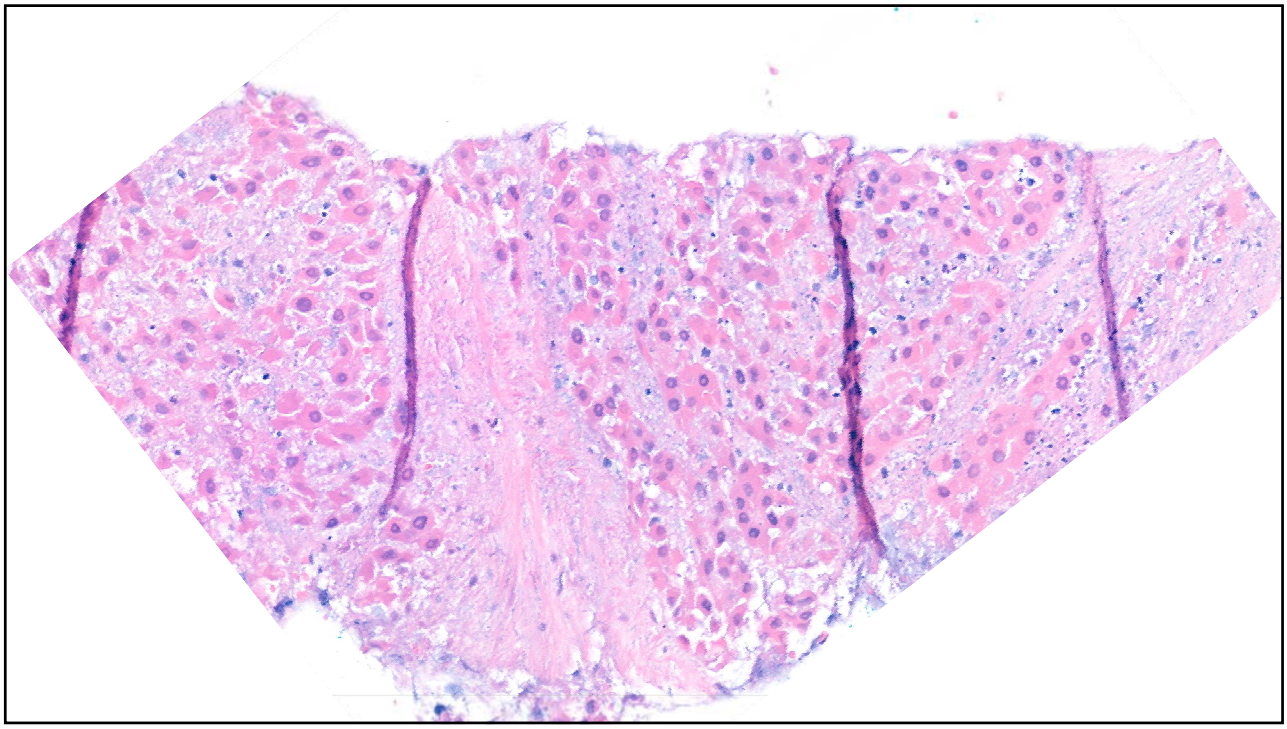
- Poorly differentiated carcinoma with hepatoid features
- Metastatic hepatoid carcinoma
- Metastatic carcinoma

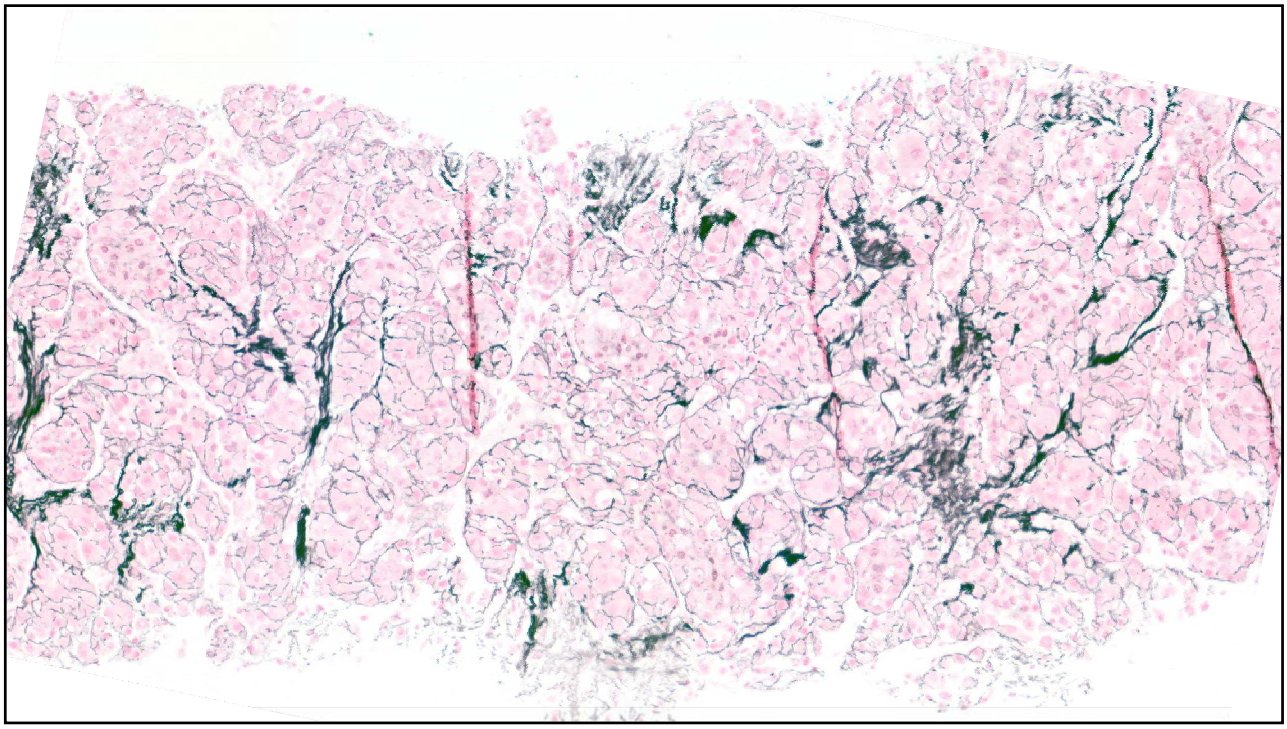
Follow-up

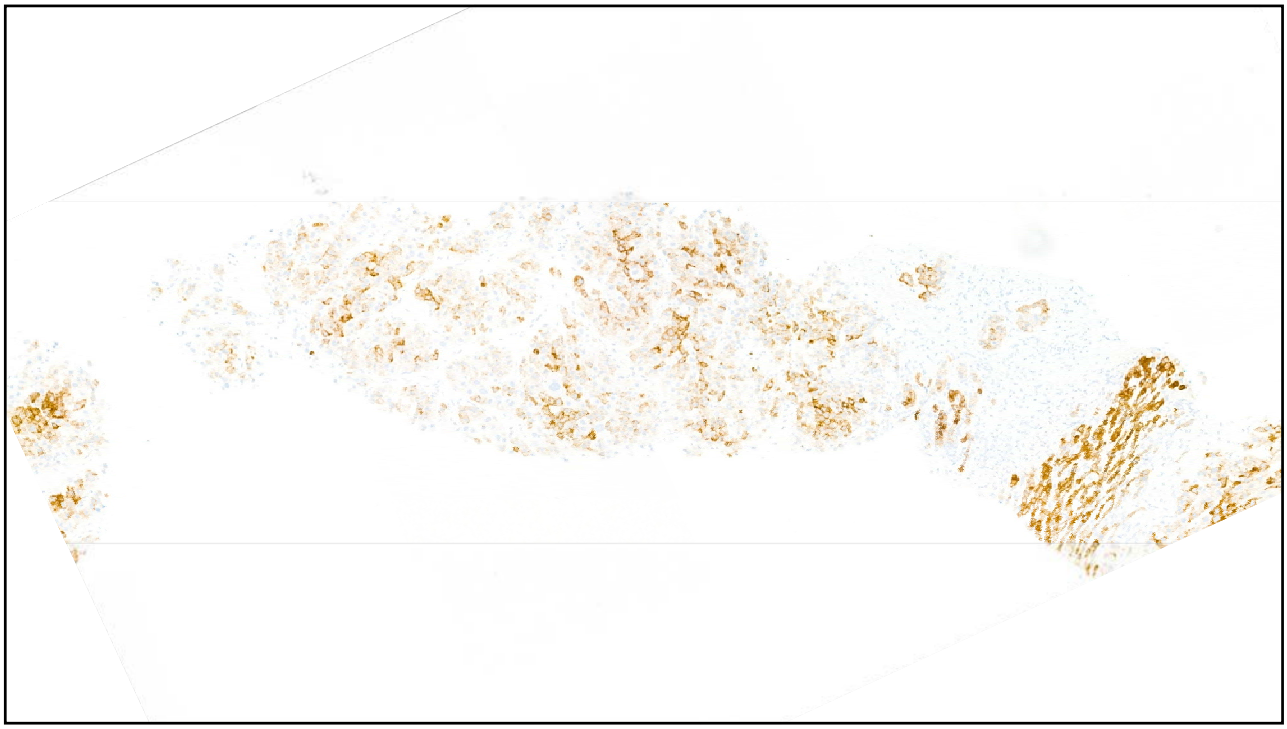
- Biopsy
- Molecular testing results

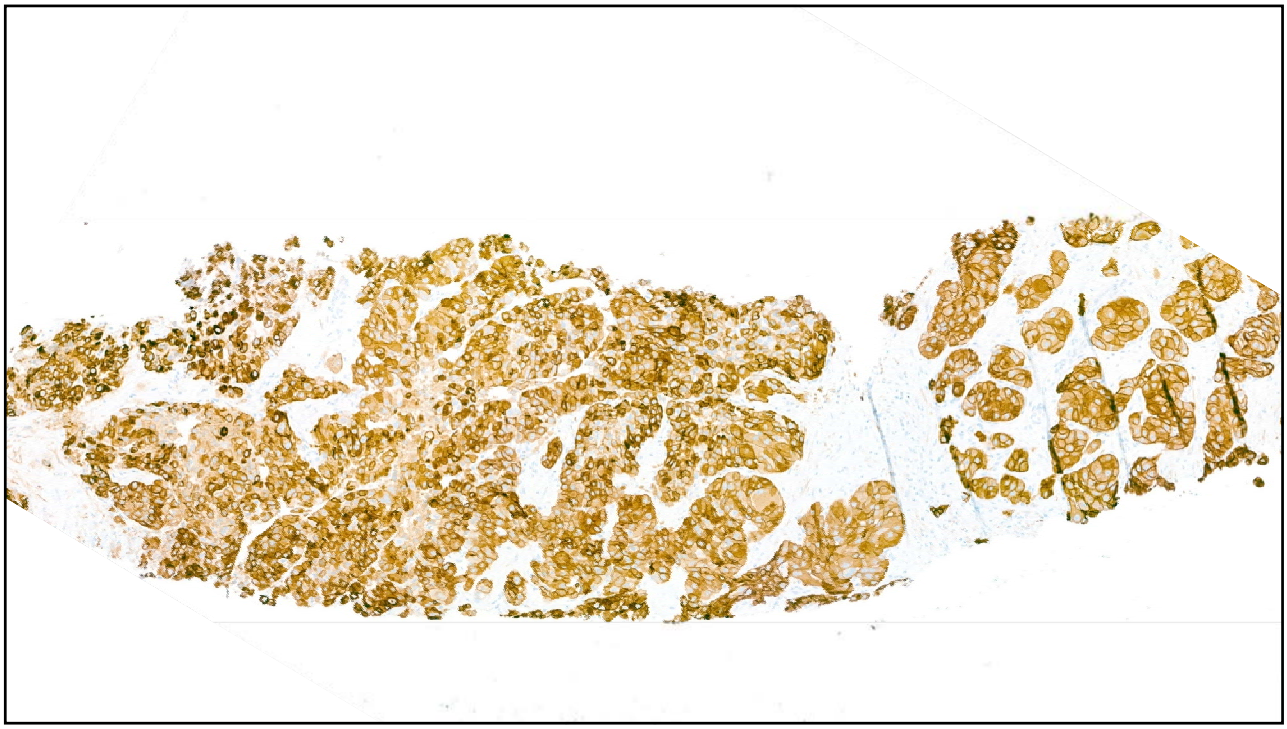












Cancer-Type Relevant Biomarkers

Biomarker	Method	Analyte	Result
DNAJB1	**Seq**	**RNA-Tumor**	**Pathogenic Fusion**
BRAF	Seq	DNA-Tumor	Mutation Not Detected
MSI	Seq	DNA-Tumor	Stable
NTRK1/2/3	Seq	RNA-Tumor	Fusion Not Detected
RET	Seq	RNA-Tumor	Fusion Not Detected
Tumor Mutational Burden	Seq	DNA-Tumor	Low, 3 mut/Mb
CCND1	CNA-Seq	DNA-Tumor	Amplification Not Detected

Biomarker	Method	Analyte	Result
CTNNB1	Seq	DNA-Tumor	Mutation Not Detected
ERBB2 (Her2/Neu)	CNA-Seq	DNA-Tumor	Amplification Not Detected
	IHC	Protein	Negative | Score 0
PD-L1 (SP142)	IHC	Protein	Negative | 0%
TERT promoter	Seq	DNA-Tumor	Mutation Not Detected

Genes Tested with Pathogenic or Likely Pathogenic Alterations

Gene	Method	Analyte	Variant Interpretation	Protein Alteration	Exon	DNA Alteration	Variant Frequency %
DNAJB1	Seq	RNA-Tumor	Pathogenic Fusion	DNAJB1-PRKACA	1	-	-

Unclassified alterations for DNA and RNA sequencing can be found in the MI Portal.
Formal nucleotide nomenclature and gene reference sequences can be found in the Appendix of this report.
Variants of Uncertain Significance can be found in the MI Portal.

Gene Fusion and Transcript Variant Detection by RNA Sequencing

GENES TESTED WITH GENE FUSION OR TRANSCRIPT VARIANT DETECTED				
Biomarker	**Fusion/Isoform**	**Splice Site**	**Transcript ID**	**Variant Interpretation**
DNAJB1	DNAJB1:PRKACA	exon 1:exon 2	NM_006145.3/NM_001304349.2	Pathogenic Fusion

Interpretation: A DNAJB1::PRKACA fusion was detected by RNA sequencing. This fusion is associated with fibrolamellar hepatocellular carcinoma (PMID: 24578576) as well as intraductal oncocytic papillary neoplasm of the pancreas/bile duct (PMID: 31678302). Exon 1 of DNAJB1 (NM_006145.3) is joined in-frame to exon 2 of PRKACA (NM_001304349.2).

Hepatocellular carcinoma (conventional)

- Transgressing capillaries
- Bare nuclei and cytoplasmic vacuolation
- Bile
- Endothelial wrapping
- Prominent nucleoli
- Nuclear holes/vacuoles
- Trabecular arrangement (cell block preparations)

WHO Classification of Tumours of the Digestive System: HCC subtypes

- Steatohepatitic
- Clear cell
- Macrotrabecular massive
- Scirrhous
- Chromophobe
- Fibrollamellar carcinoma
- Neutrophil-rich
- Lymphocyte-rich

Fibrolamellar Carcinoma

Clinical follow-up

- On gemcitabine/oxaliplatin + Lenvatinib

ADVANCES IN CYTOLOGY AND SMALL BIOPSIES – VIRTUAL MICROSCOPY SESSION

Case #3:
71 y/o postmenopausal woman,
routine screening

Jeffrey Mito MD PhD
6/9/2025

History

- Recent “abnormal” pap at an outside hospital

Diagnosis?

- A) NILM-R
- B) Atypical glandular cells, NOS
- C) Atypical glandular cells, favor neoplastic
- D) Adenocarcinoma, NOS
- E) Other

Final Cytologic Diagnosis

Satisfactory for evaluation; transformation zone present.

EPITHELIAL CELL ABNORMALITY - GLANDULAR.
Atypical glandular cells, favor neoplastic.

The Bethesda System: Glandular Abnormalities

- Atypical glandular cells, not otherwise specified (NOS)
  - If possible, specify endocervical or endometrial
- Atypical glandular cells, favor neoplastic
  - Specify if endocervical
- Endocervical adenocarcinoma *in situ* (AIS)
- Adenocarcinoma
  - Endocervical, endometrial, extrauterine, or not otherwise specified

Atypical Glandular Cell – Favor Neoplastic

- Cell morphology either quantitatively or qualitatively falls short of a diagnosis of *in situ* or invasive adenocarcinoma
- Higher rate of invasive cancer and significant pathology vs AGC-NOS: 55-65% vs 24-32%
- In one study, most predictive of glandular neoplasia on histologic follow-up (21 of 26, 81%)

Chatchotikawong U, et al. Int. J. Gyn. Obst. (2012) **119**: 30
Pradhan D., et al. Can. Cyto. (2016) **124:** 589

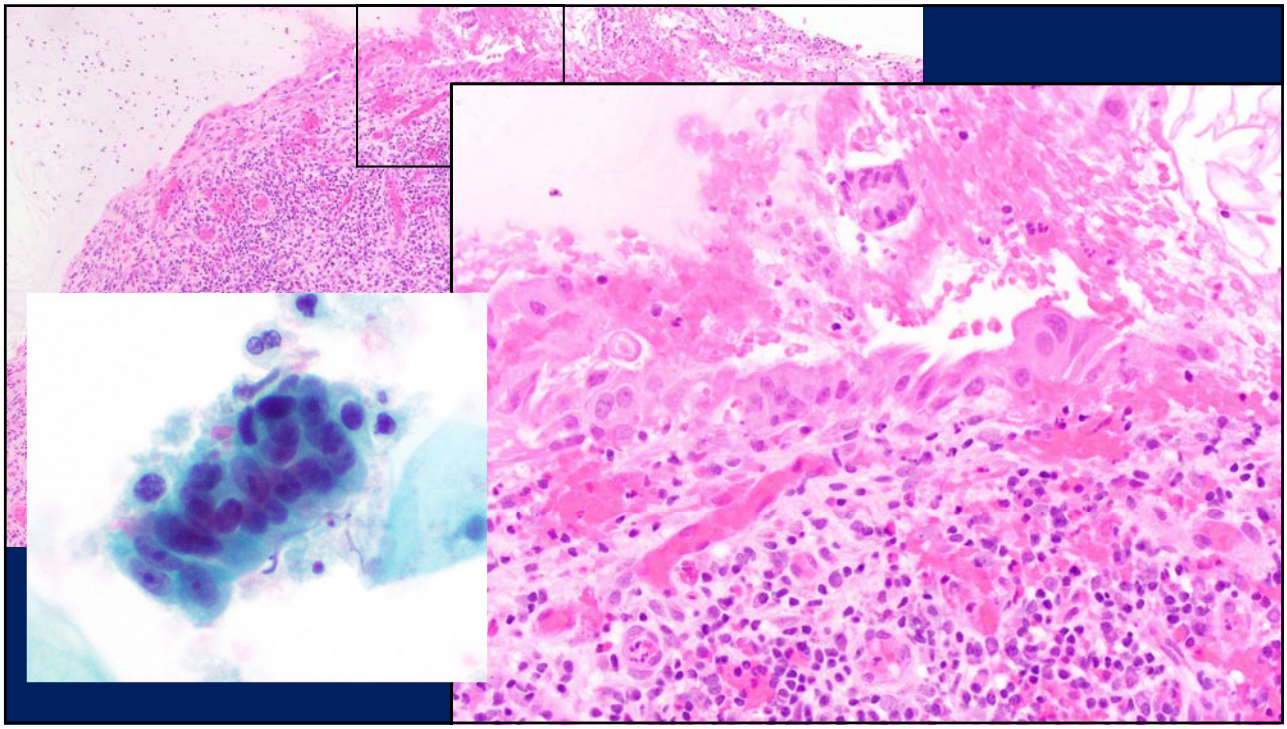
ASCCP Management Guidelines

- All subcategories of "Atypical glandular cells" (except "Atypical endometrial cells") should be managed with colposcopy and endometrial sampling (if $\geq$ 35 years of age or age <35 and at risk for endometrial neoplasia)
  - Abnormal uterine bleeding
  - Obesity
  - Chronic anovulation
  - Family history or genetic predisposition

Perkins RB, et al. J. Low. Gen. Tract. Dis. (2020) **24**: 102

Follow-up

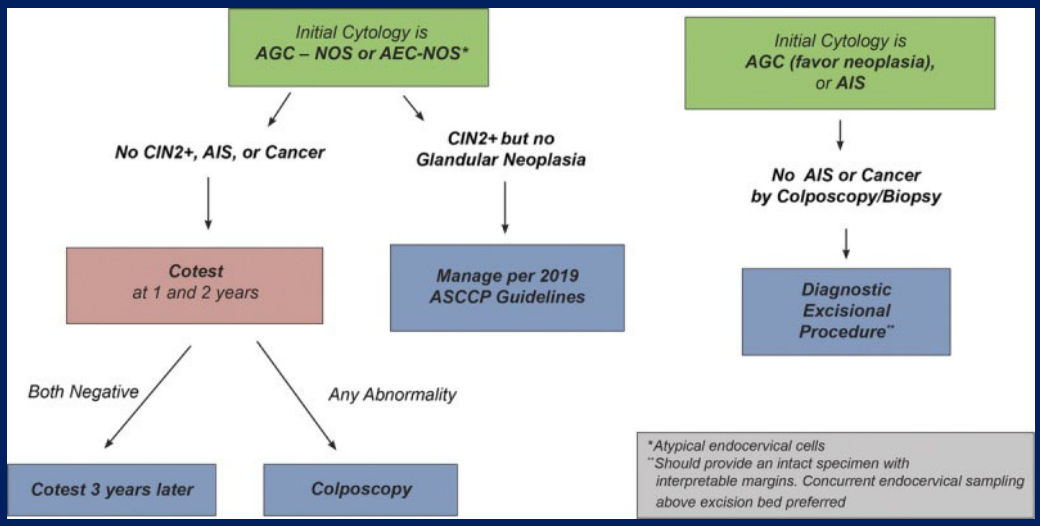
- Follow-up endometrial sampling was benign



Endocervical Polyps

- Similarities to adenocarcinoma:
  - Nuclear enlargement
  - Prominent nucleoli
  - Mitotic activity
- Lack of necrotic debris
- Lack of isolated single cells
- Correlation with clinical exam

ASCCP Management Guidelines



Perkins RB, et al. J. Low. Gen. Tract. Dis. (2020) **24**: 102

Follow-up

- Follow-up endocervical sampling, pap test, and HR-HPV testing were all negative for the subsequent two years
- Patient is alive and well 10+ years later

Take Home Points

- Reparative changes can mimic invasive cancer
- History and clinical exam can refine the differential of an AGC diagnosis
- Specifying endometrial or endocervical atypia is important for subsequent patient management

Questions?

ADVANCES IN CYTOLOGY AND SMALL BIOPSIES – VIRTUAL MICROSCOPY SESSION

Case #4:
71 y/o postmenopausal woman,
routine screening

Jeffrey Mito MD PhD
6/9/2025

History

- No prior abnormal pap tests
- No screening in the last 20 years
- Normal exam

Diagnosis?

- A) HSIL
- B) Squamous cell carcinoma
- C) Atypical glandular cells, NOS/favor neoplastic
- D) Adenocarcinoma *in situ*
- E) Other

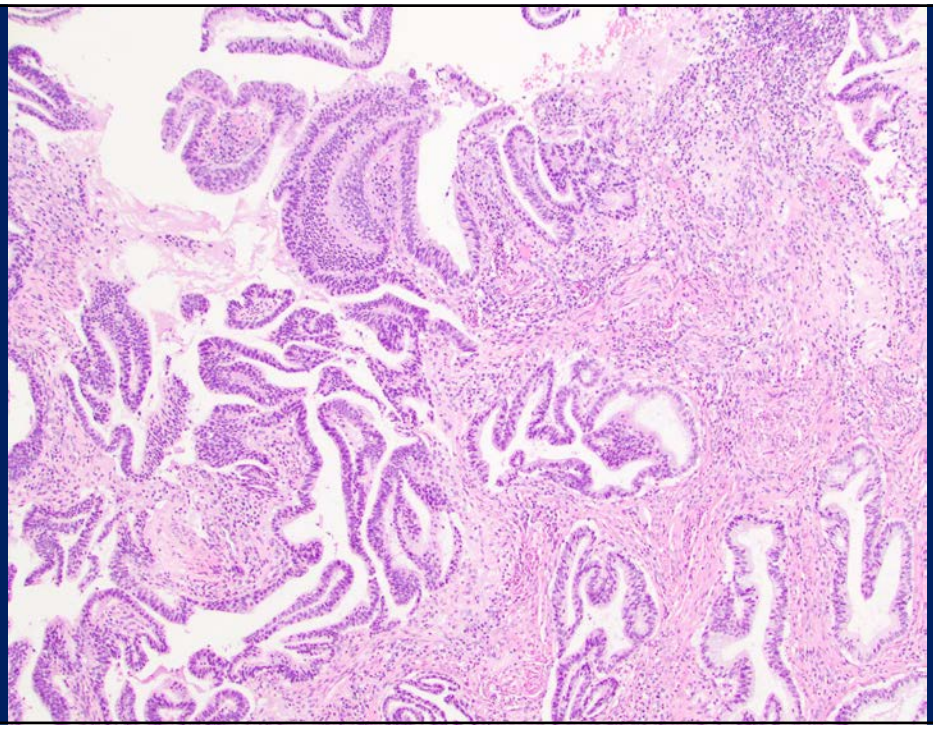
Final Cytologic Diagnosis

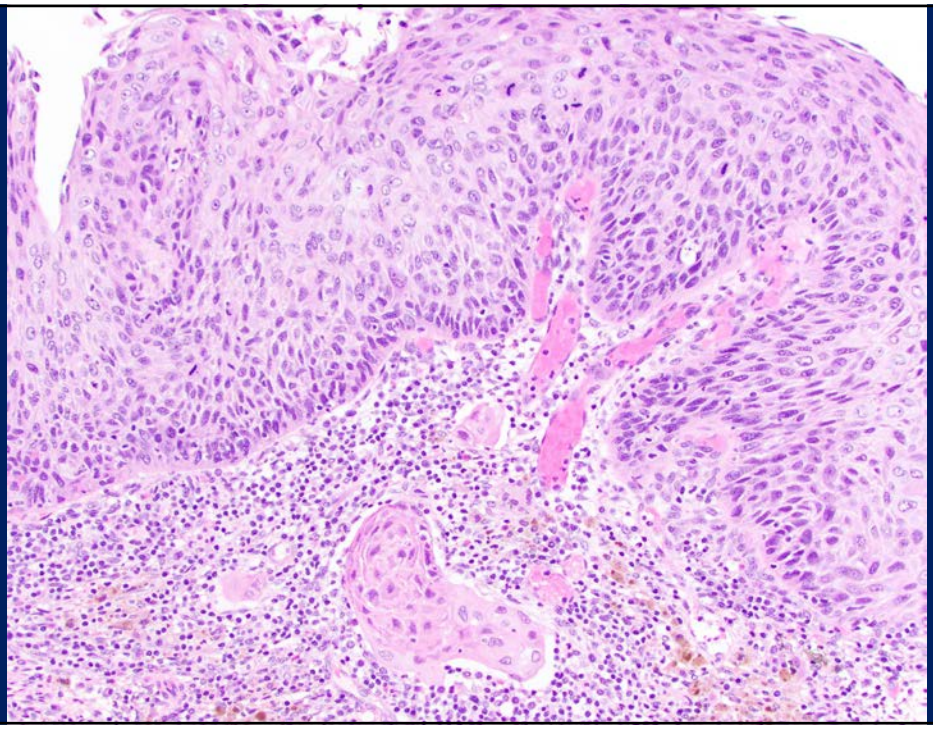
Satisfactory for evaluation; transformation zone present.

EPITHELIAL CELL ABNORMALITY - GLANDULAR.
Adenocarcinoma, not otherwise specified.

EPITHELIAL CELL ABNORMALITY - SQUAMOUS.
High grade squamous intraepithelial lesion (see NOTE).

NOTE: The presence of a more significant lesion cannot be excluded.





Final Pathologic Diagnosis

INVASIVE ENDOCERVICAL ADENOCARCINOMA, MUCINOUS (HPV-associated) TYPE

HIGH GRADE SQUAMOUS INTRAEPITHELIAL LESION (CIN3) involving endocervical crypts, with one focus representing superficial invasion (<1 mm)

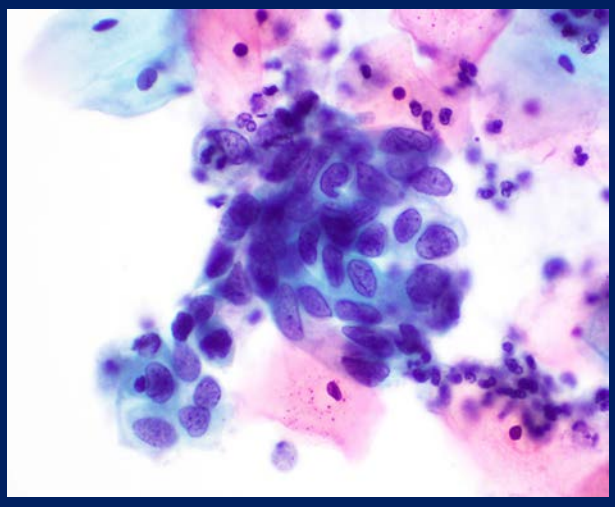
Squamous and Glandular Abnormalities on Pap Test

- Identifying BOTH squamous and glandular abnormalities on Pap test is infrequent:
  - <0.1% (230 of 361,953) of Pap tests
- Histologic follow-up:
  - Squamous lesions: 52-53%
  - Glandular lesions: 6-7%
  - Both squamous and glandular lesions: 3-4% (AIS+SIL)
- The majority are positive for HR-HPV (57-66%)

Yan Khor L, et al. Can. Cyto. (2014) **122**: 620
Harbhajanka A, et al. Diag. Cyto. (2019) **47**: **88**

HSIL with Gland Involvement

- Features:
  - Cells with centrally whorled or spindled cells and peripheral flattening
  - Clusters or sheets of atypical cells with pseudostratification and peripheral palisading (mimicking AIS)



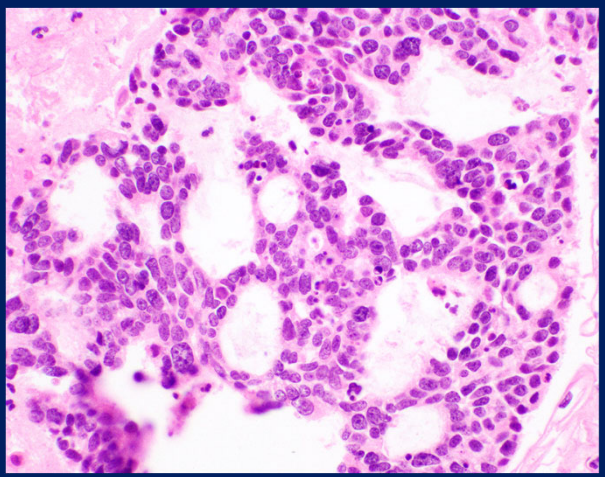
HSIL with Gland Involvement

- Interobserver agreement of HSIL with gland involvement on pap test is poor
- Histologic identification of HSIL with endocervical gland involvement has been linked to increased rates of detection of HSIL (32 vs 25%) or ASC-H (17 vs 12%) on pap test
- Features favoring HSIL over AIS
  - Limited feathering, rosettes or columnar cells
  - Denser cytoplasm
  - Single atypical squamous cells in the background

Jones R, et al. JASC (2020) **9**: 137

The Utility of Cell Blocks

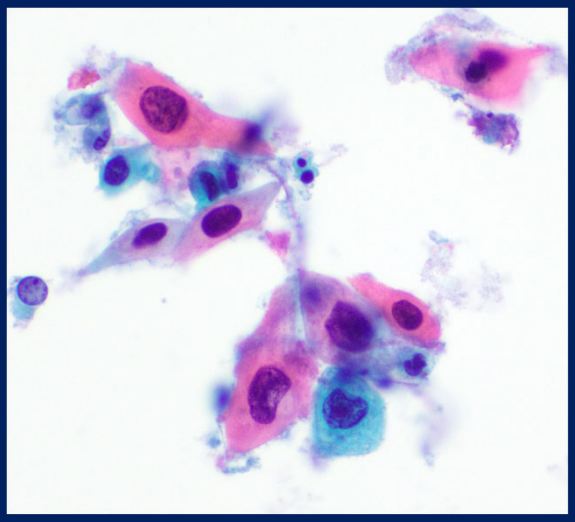
- Cell blocks can be useful in the differential diagnosis of Atypical glandular cells:
  - Xing, et al.: 148 patients with pap tests screened as AGC
    - 31 of 68 (46%) samples screened as AGC reclassified as NILM/LSIL
    - 7 of 68 (10%) were given a specific diagnosis: HSIL, AIS, or invasive ACA



Xing W, et al. Can. Cyto. (2014) **122**: 8

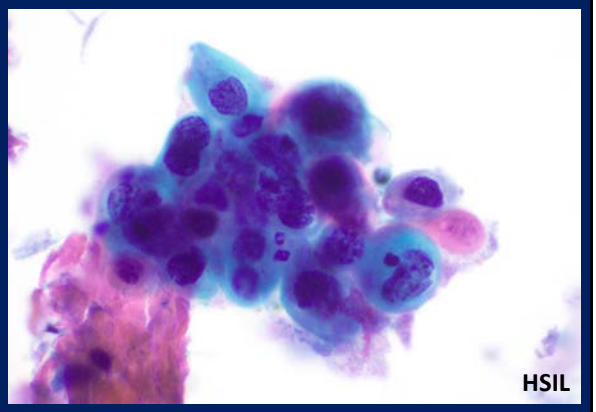
Cytologic Criteria for Squamous Cell Carcinoma

- Cells occur singly or in groups with poorly defined cell borders
- Keratinizing SQC: typically isolated single cells or aggregates of markedly variable cell size and shape
- Display most of the features of HSIL
- Nuclei with markedly irregular clumped or coarse chromatin
- Nucleoli may be prominent
- Tumor diathesis



HSIL vs Invasive Squamous Cell Carcinoma

- Challenging to differentiate:
  - Not all invasive cancers will have a tumor diathesis – especially true with liquid based cytology
  - Tumor diathesis can be seen in benign conditions and HSIL
  - Keratinizing SILs can have highly abnormal keratinized cells
  - Prominent nucleoli can be seen in HSIL
- False positives often caused by a constellation of these findings



Take Home Points

- The diagnosis of both a squamous and glandular abnormality on Pap test is rare - most of these cases will represent HSIL on histologic follow-up
- The distinction between HSIL and squamous cell carcinoma can be challenging
- Cell blocks can be helpful to further characterize hyperchromatic crowded groups and refine a diagnosis of AGC

Questions?