UCSF
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WELL-DIFFERENTIATED HEPATOCYTIC TUMORS: *UPDATE AND DIAGNOSTIC PROBLEMS*

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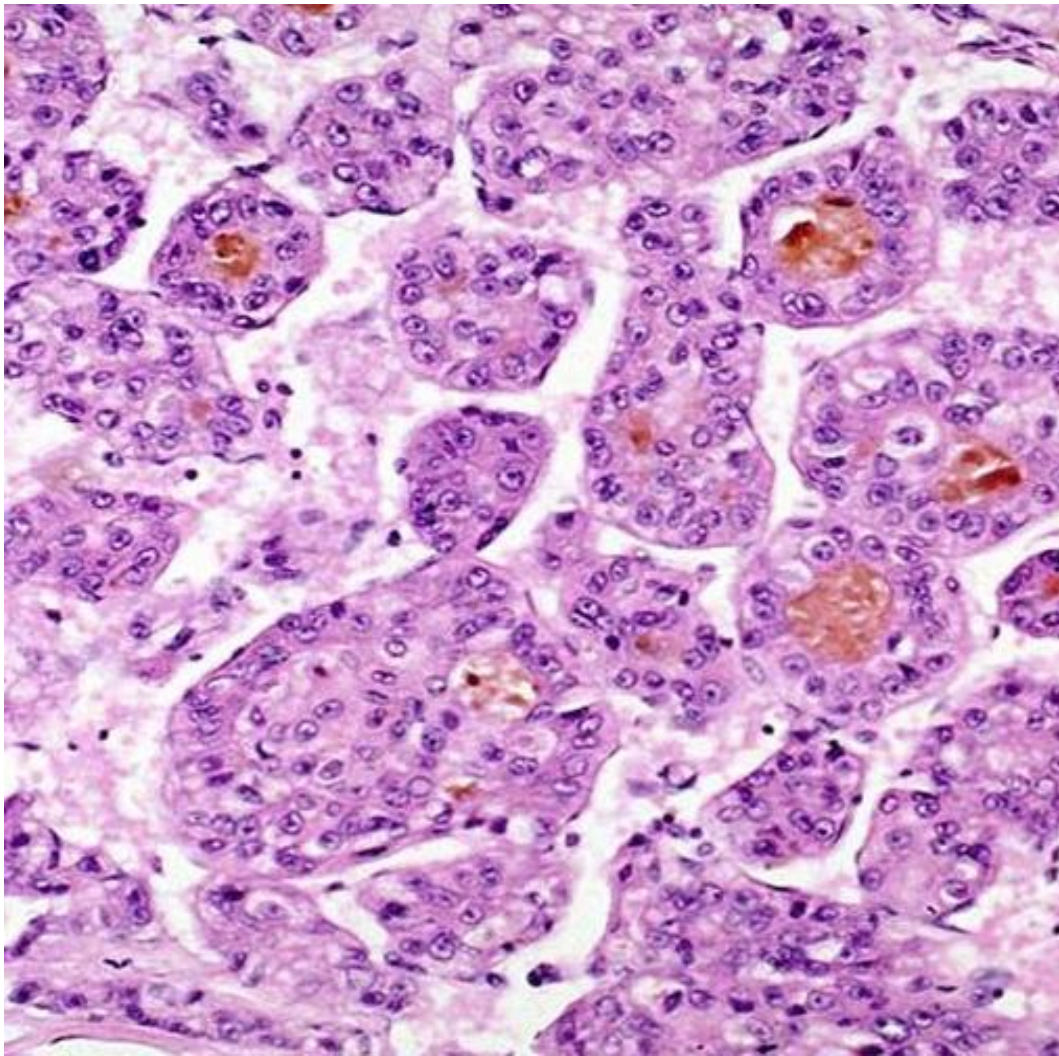
Distinction of Hepatic Tumors in the Liver

- Most diagnoses of Hepatocellular Carcinoma (HCC) not a problem
- Example:
  - 6 cm mass in a patient with HCV *CIRRHOSIS*
  - elevated AFP > 1000
  - Typical histology



Important Factor: Cirrhosis or not

- Cirrhosis: Almost always HCC
  - <3 cm: possible dysplastic nodule (DN)
  - 5-10% combined HCC/cholangiocarcinoma (CC)
  - <<5% other (primary or metastatic)
- Non-Cirrhotic liver: Well-differentiated lesions
  - Hepatocellular adenoma (HCA)
  - Focal nodular hyperplasia (FNH)
  - Well-differentiated HCC

"Outstanding" nodules in cirrhosis (but not HCC)

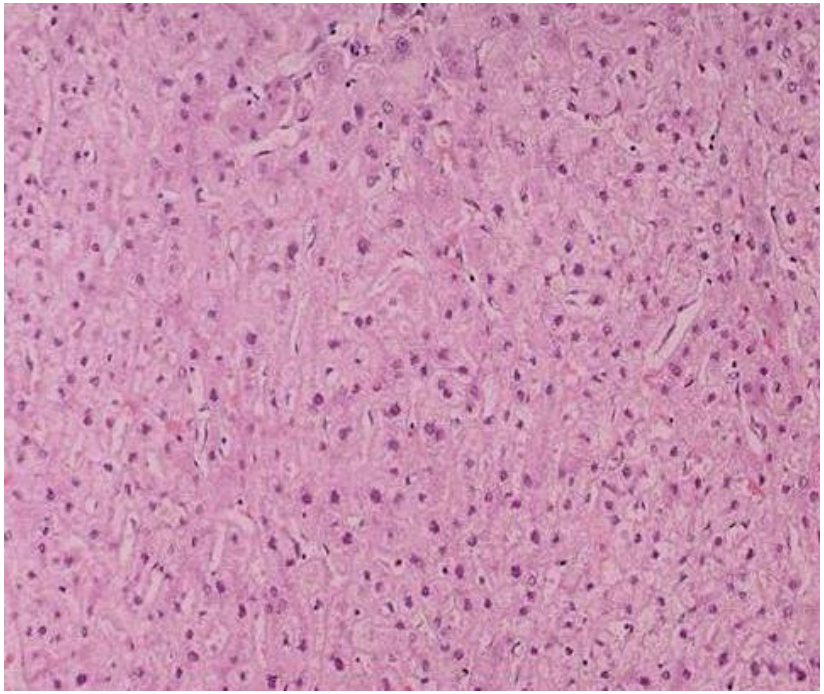
- **Macroregenerative (MRN)**
- **Dysplastic, Low Grade (LGDN):** ***Mild atypical features suggesting clonality (included in MRN category)***
- **Dysplastic, High Grade (HGDN):** ***Moderate, severe atypical features, but not diagnostic of HCC***

Macroregenerative Nodule
Low Grade Dysplastic Nodule

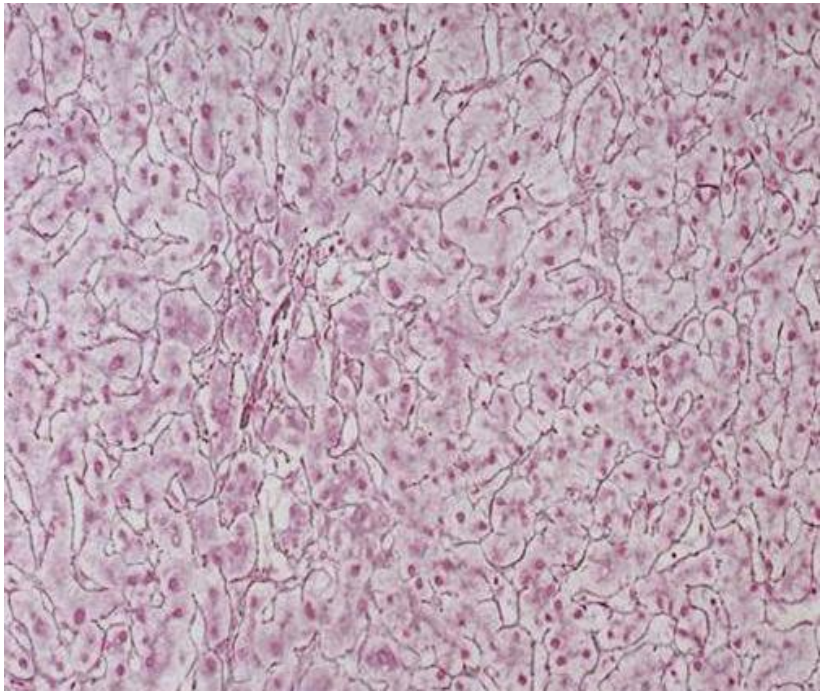
- Occurs in cirrhotic liver
- Approx. 0.8 cm to 3 cm diameter
- Usually contains portal tracts
- Histology like cirrhotic nodule
- No significant N:C changes
- Can see multiple MRNs in same liver

Macroregenerative Nodule
Low Grade Dysplastic Nodule

Bland cytology, uniform



Reticulin framework intact

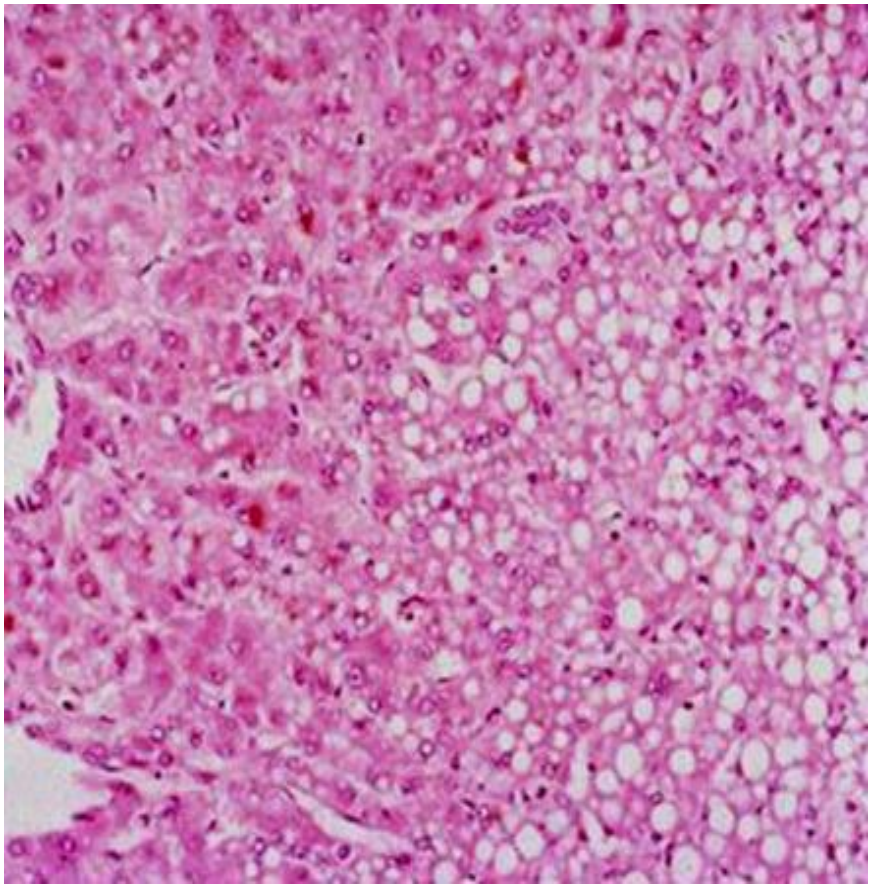


Macroregenerative Nodule
Low Grade Dysplastic Nodule

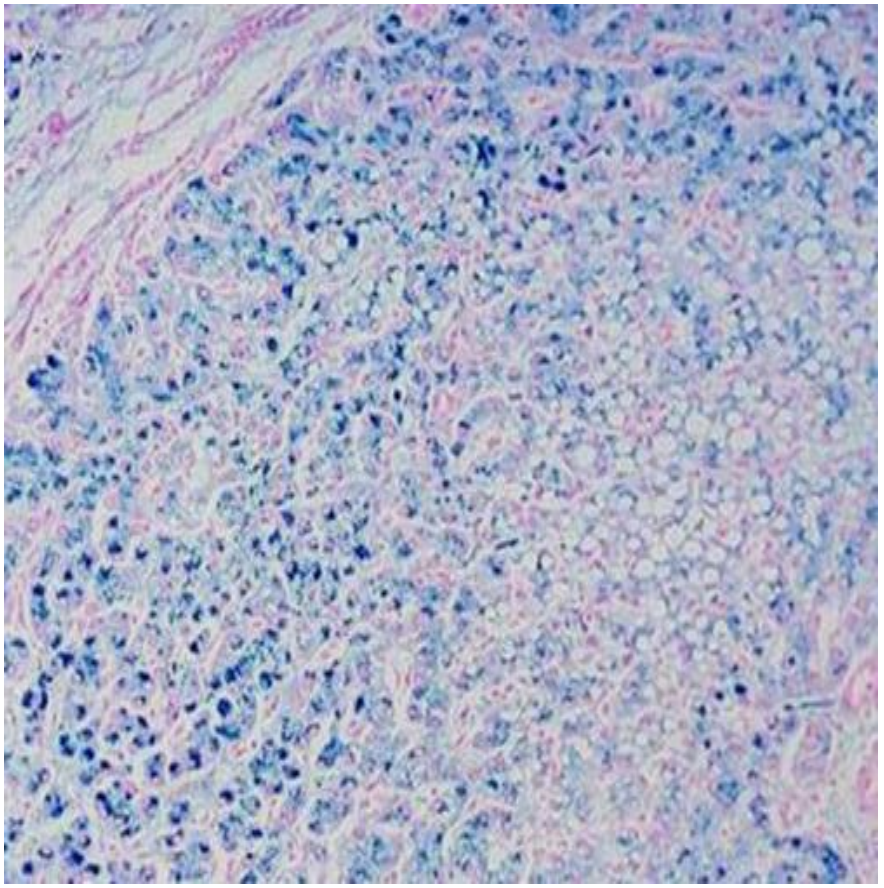
- May also contain iron, bile, Mallory's hyaline, fat, clear cell change

Macroregenerative Nodule
Low Grade Dysplastic Nodule

Macroregenerative Nodule
Low Grade Dysplastic Nodule



Fatty change



IRON STAIN: Positive

High Grade Dysplastic Nodule (HGDN)

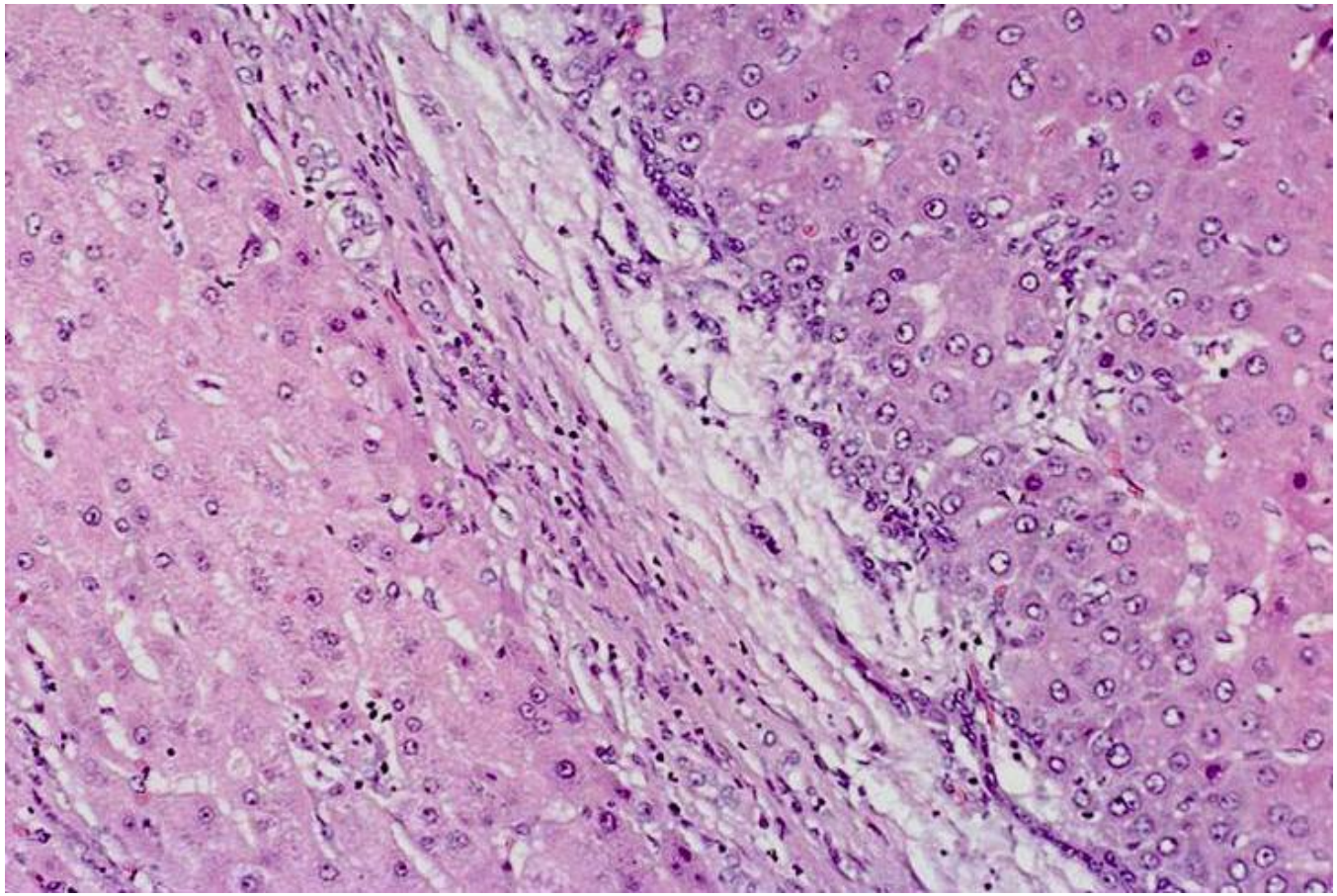
- May contain:
  - Focal decrease or absence of reticulin
  - Increased N/C ratio
  - Cell plates up to 3 cells thick, but not arranged in groups of trabeculae

High Grade Dysplastic Nodule (HGDN)

HGDN:

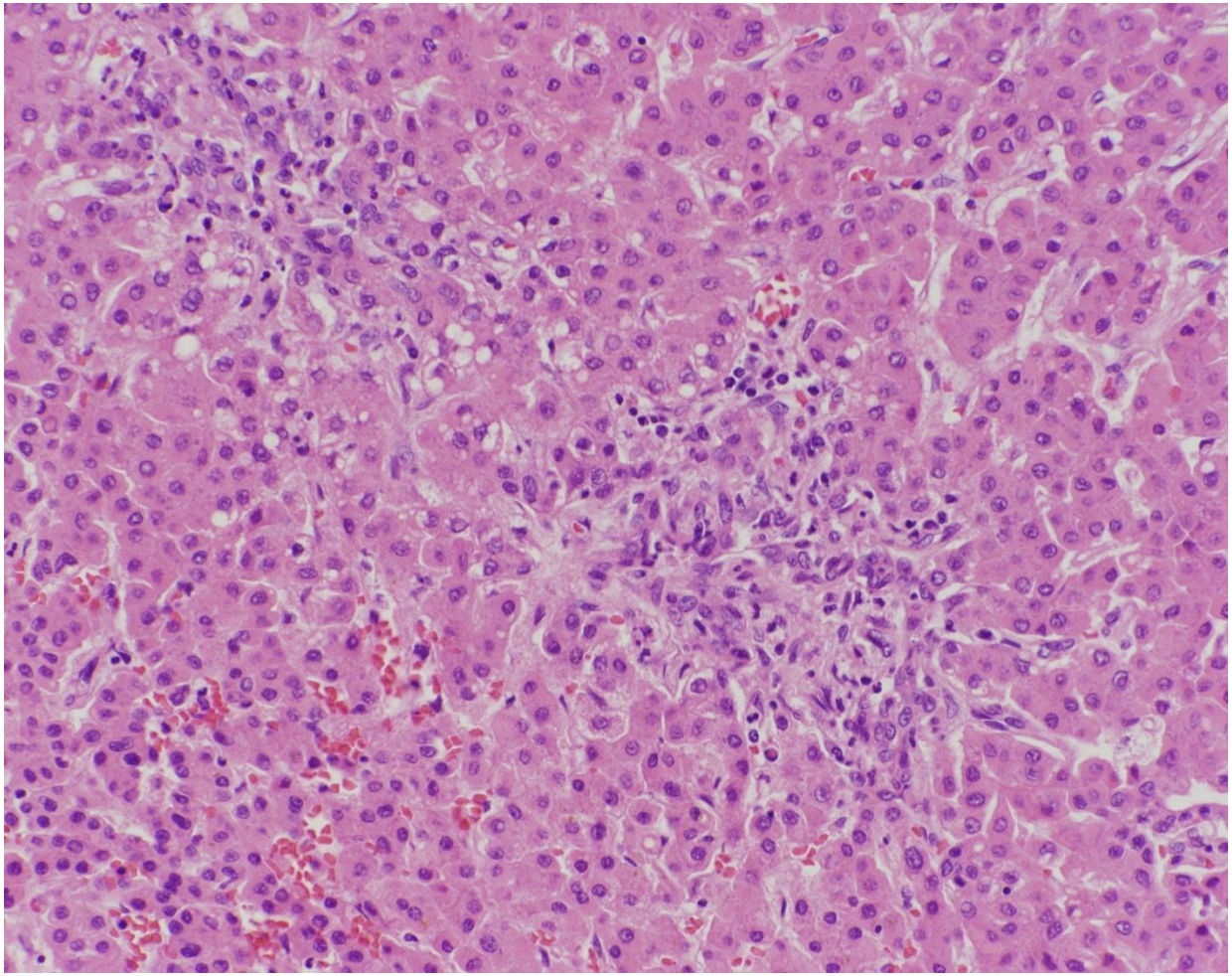
increased nuclear size and crowding

(on right)



High Grade Dysplastic Nodule (HGDN)

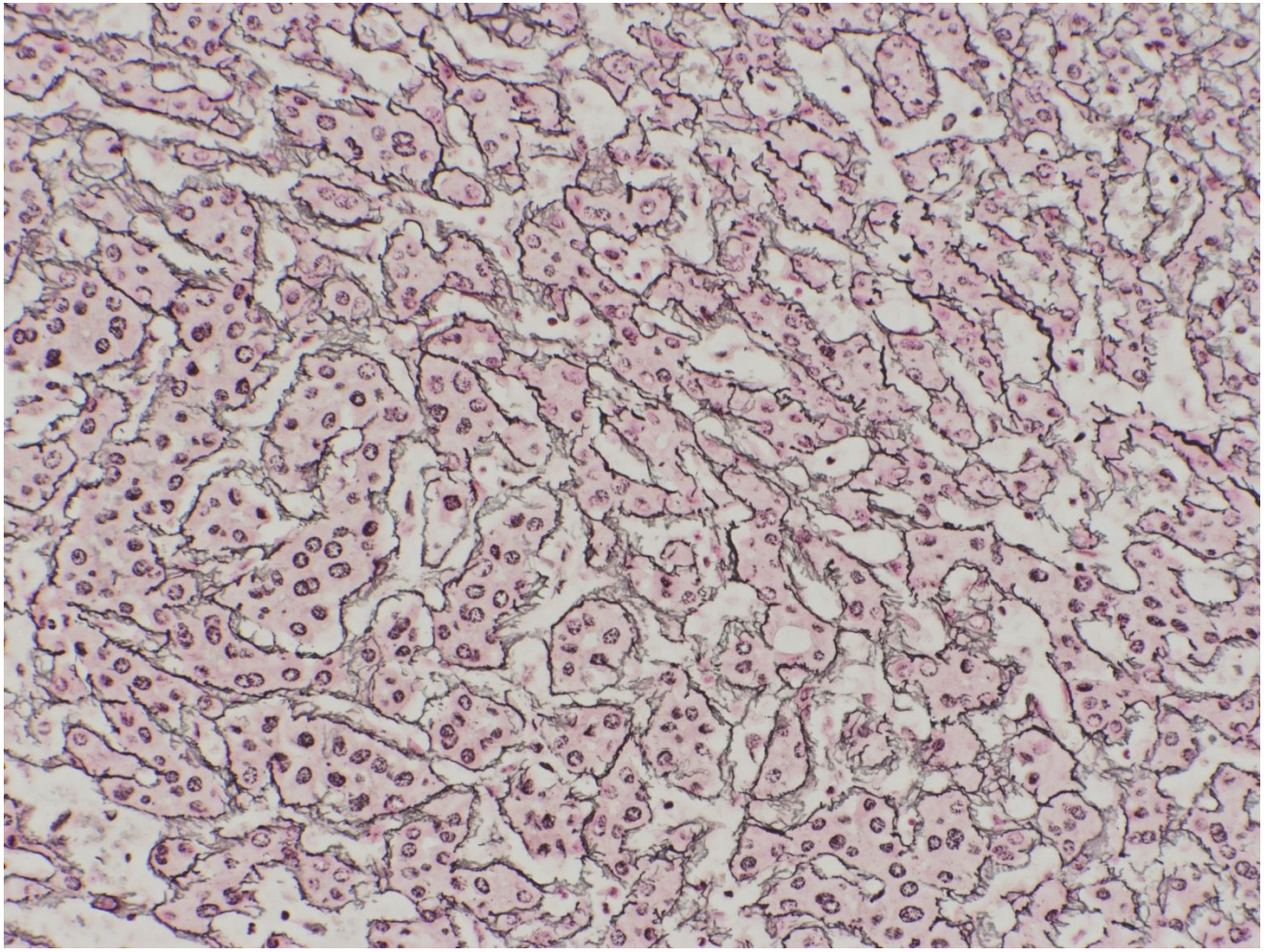
- Small cell change
- Mildly thick cell plates
- Ductular reaction



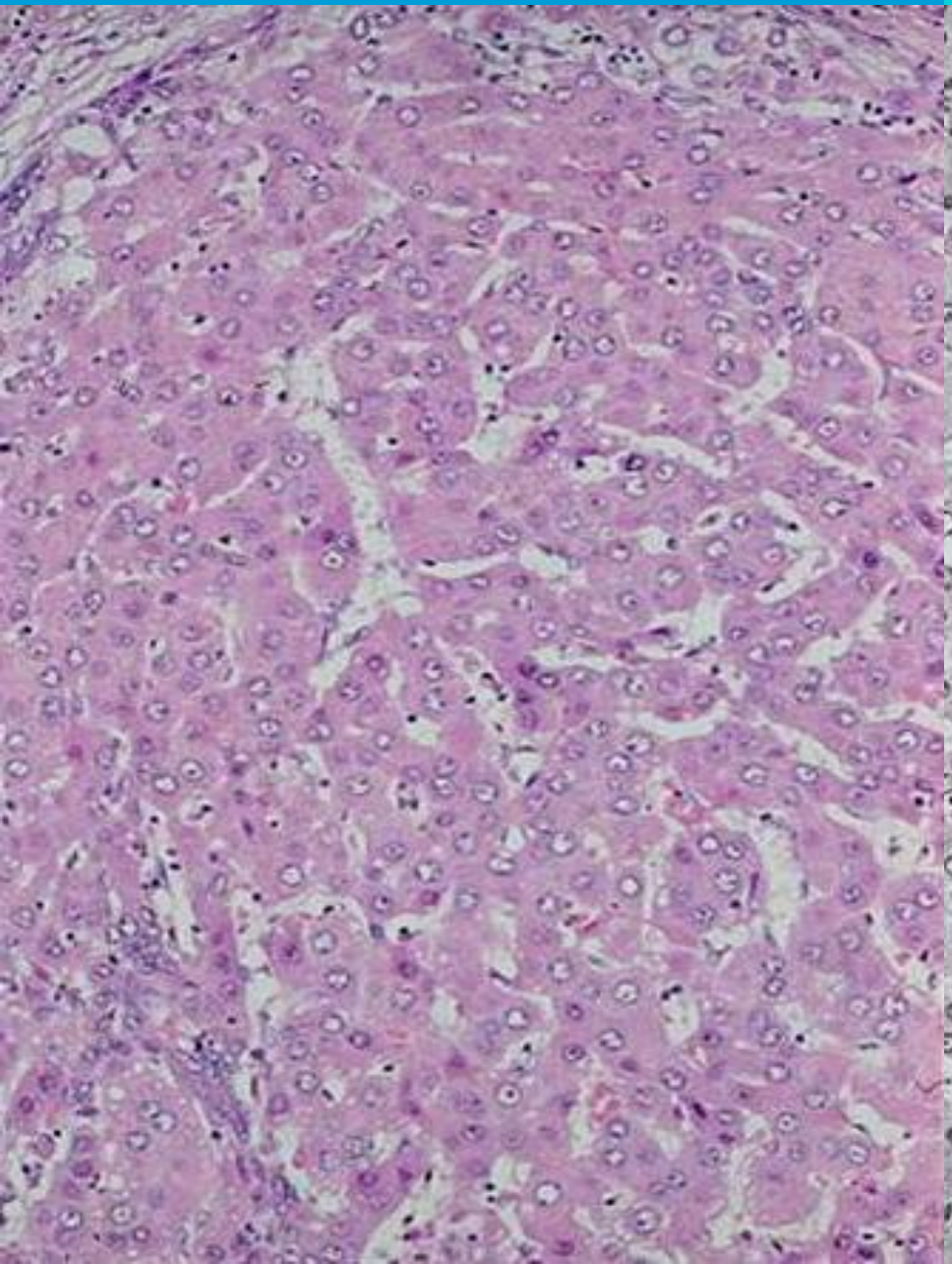
High Grade Dysplastic Nodule

RETICULIN

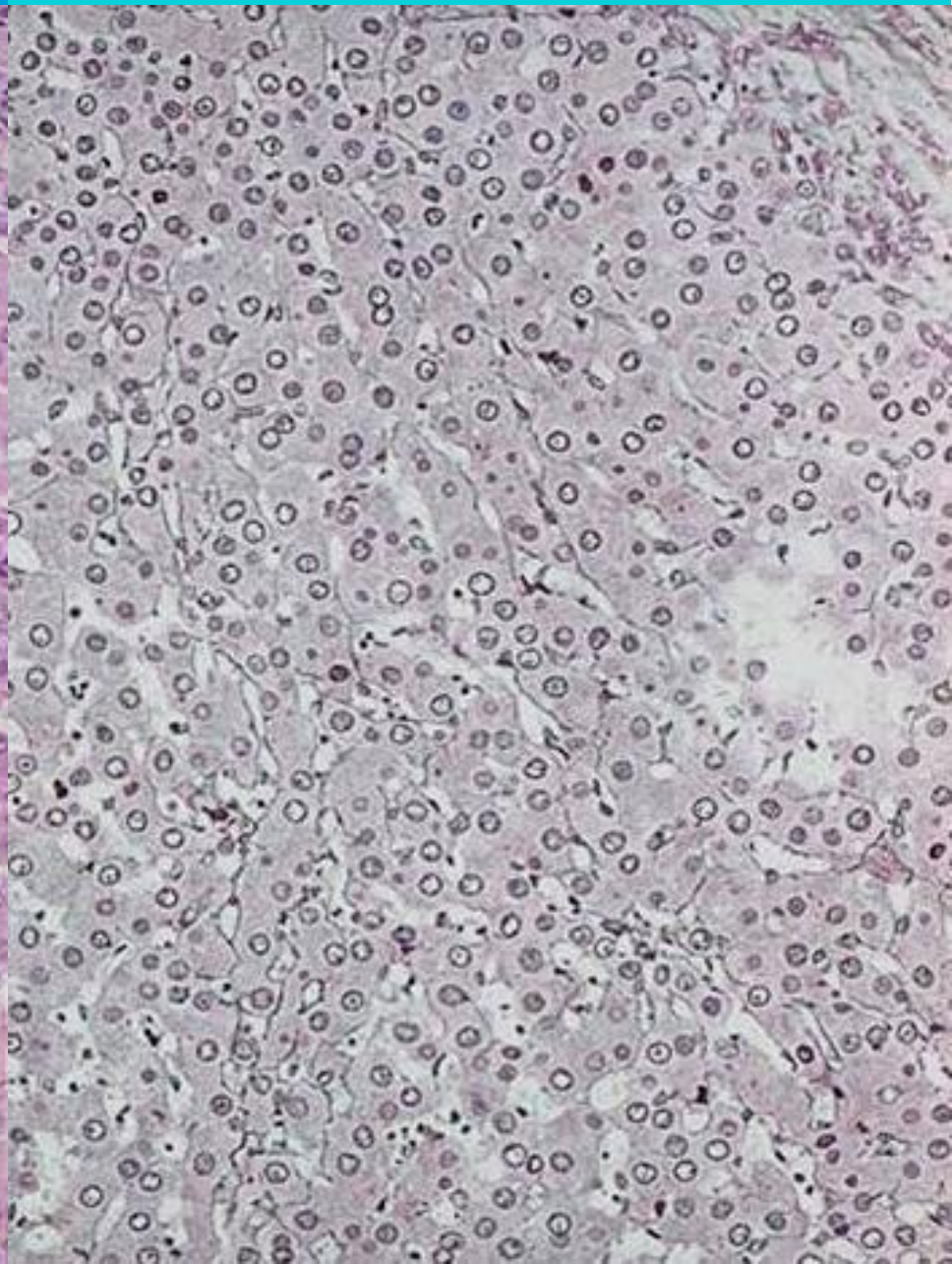
Focal, thick cell plates and small cell change



HGDN: Intact plate pattern



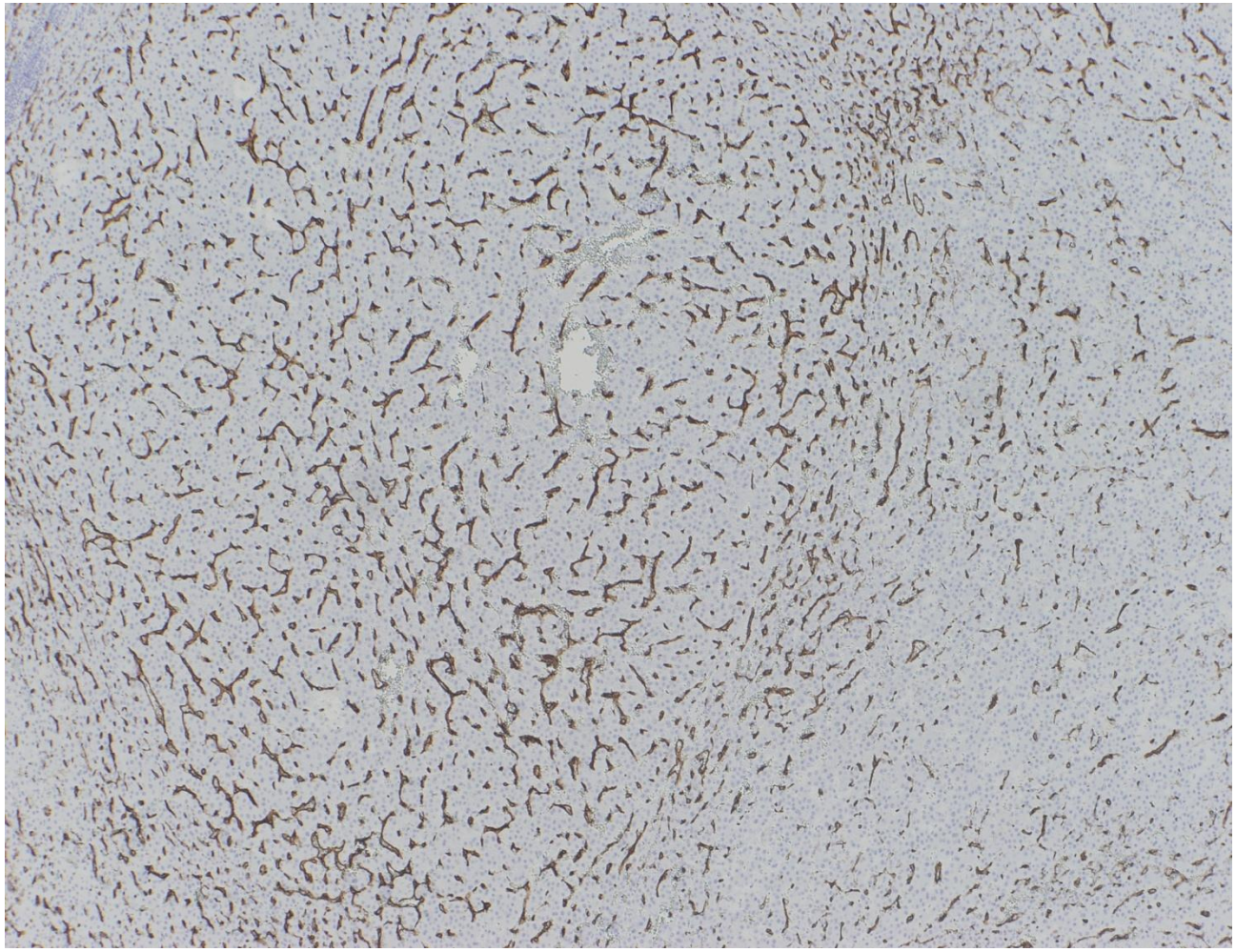
RETICULIN: Focal mild loss



High Grade Dysplastic Nodule

CD34:

Usually Increased



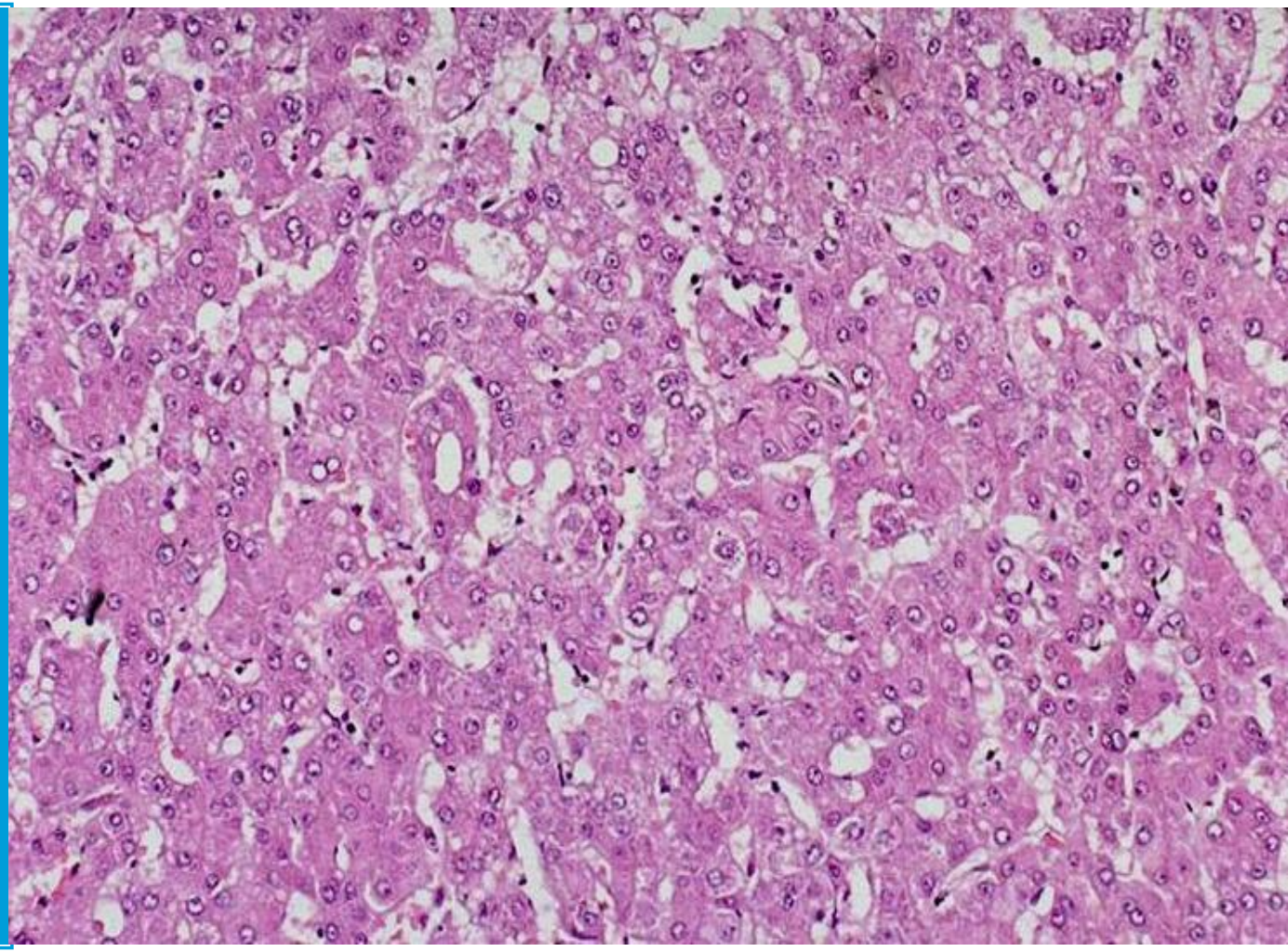
High Grade Dysplastic Nodule

May also contain:

- Bile, iron, Mallory hyaline, fat, clear cell change
- Pseudoglands may be present

High Grade Dysplastic Nodule

- Pseudogland
- Increased nuclear size and density
- Cell plate pattern intact



Small HCC/Early HCC

Lesions ≤ 2 cm

Prior to 2008, no consensus:

Criteria differed from region to region

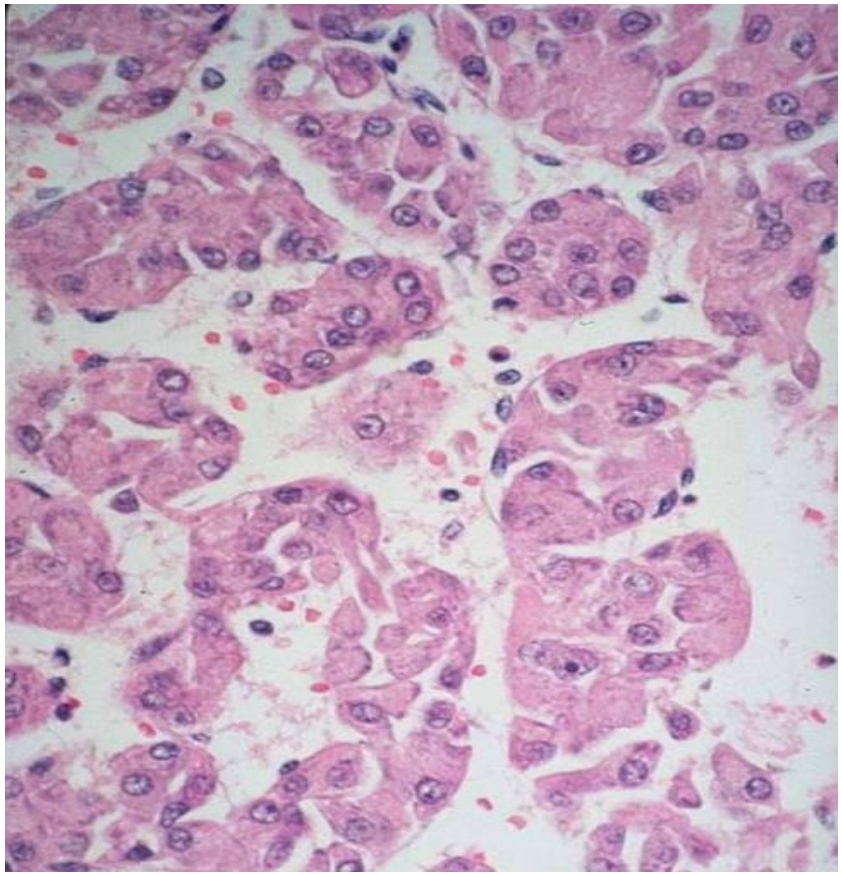
Needed to be standardized

WDHCC (small or early HCC): Histologic Features

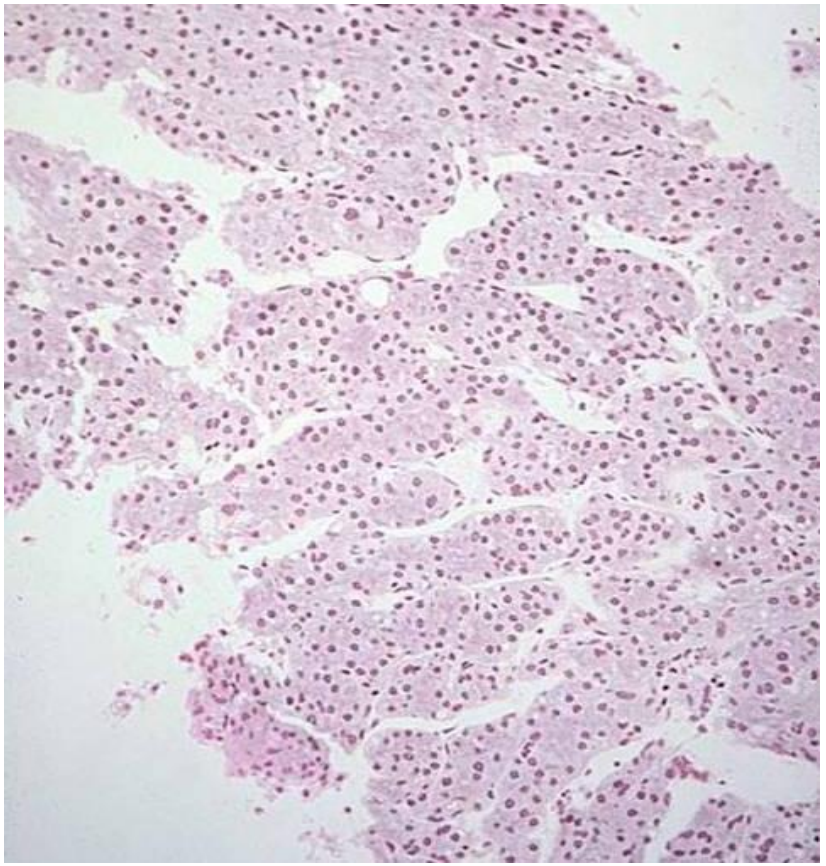
- May contain:
  - Uniformly thick cell plates > 3 cells in groups of 3 plates
  - Increased N/C ratio
  - Decreased or absent reticulin (some cases with fibrous stroma)

Well-Differentiated HCC

H&E: "Floating Plates"

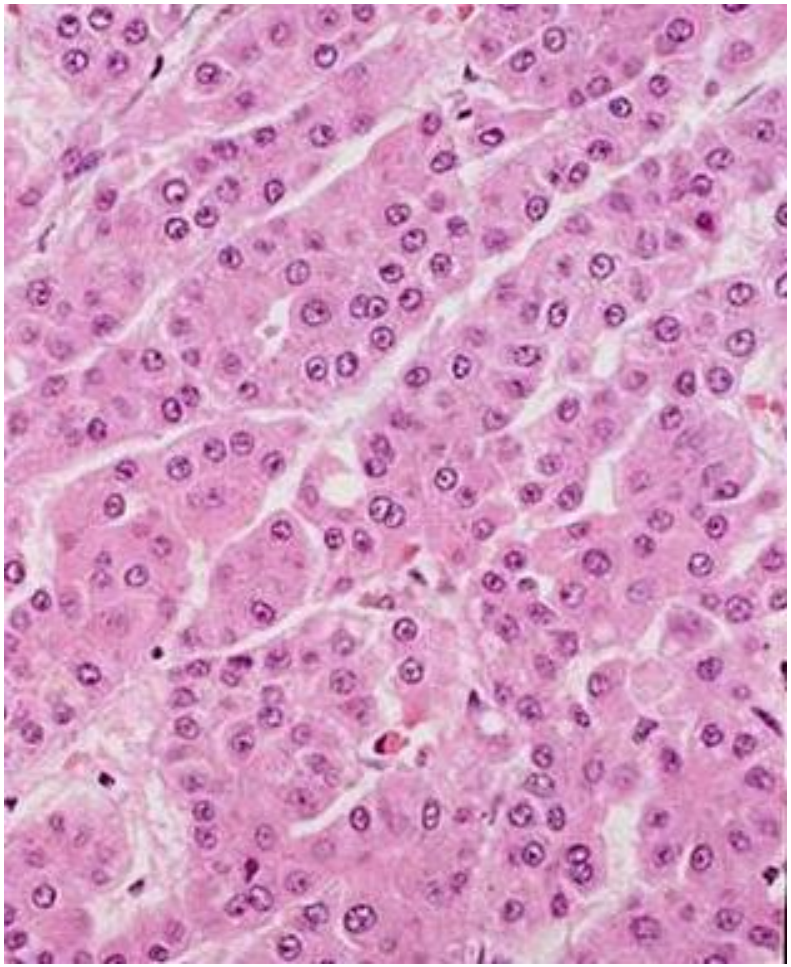


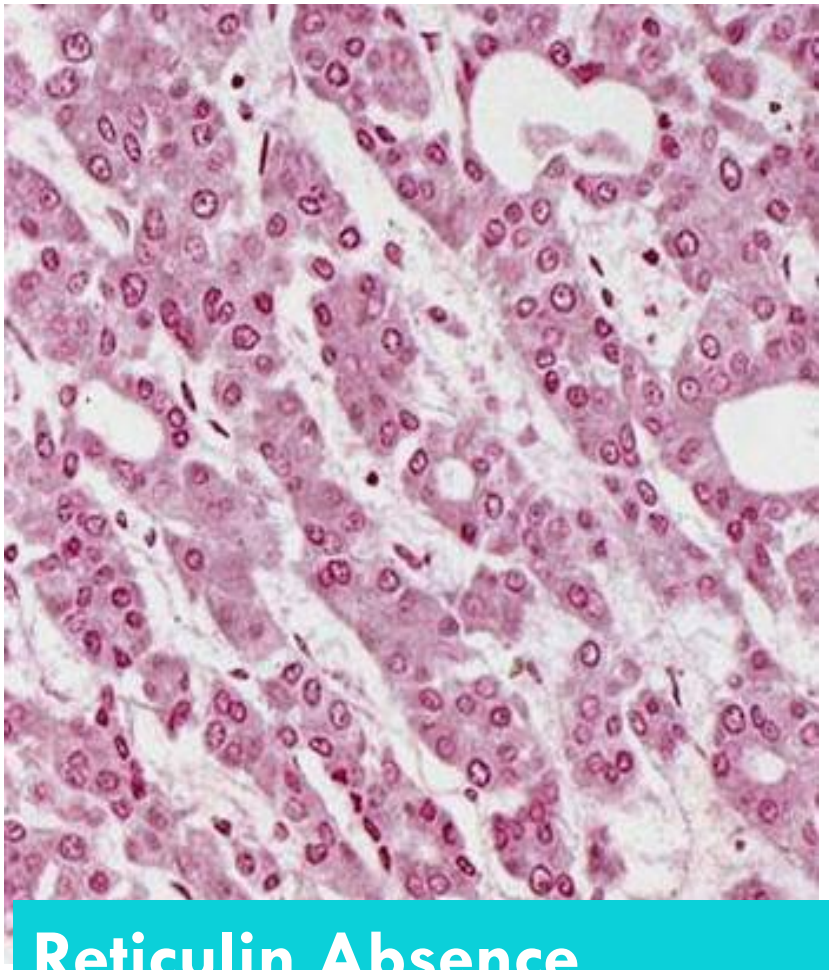
Reticulin Absence



Well-Differentiated HCC:

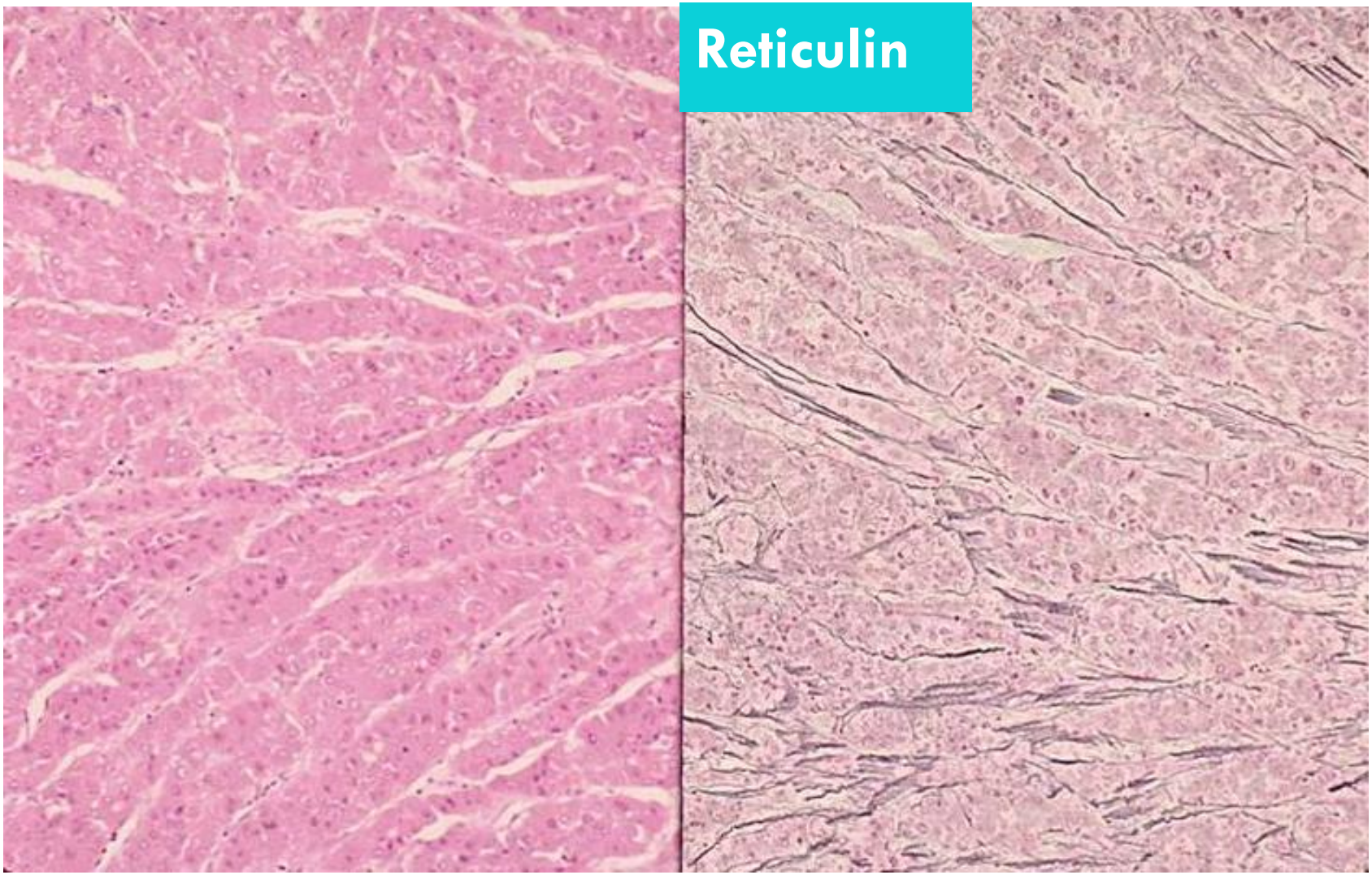
Thin, ribbon-like plates





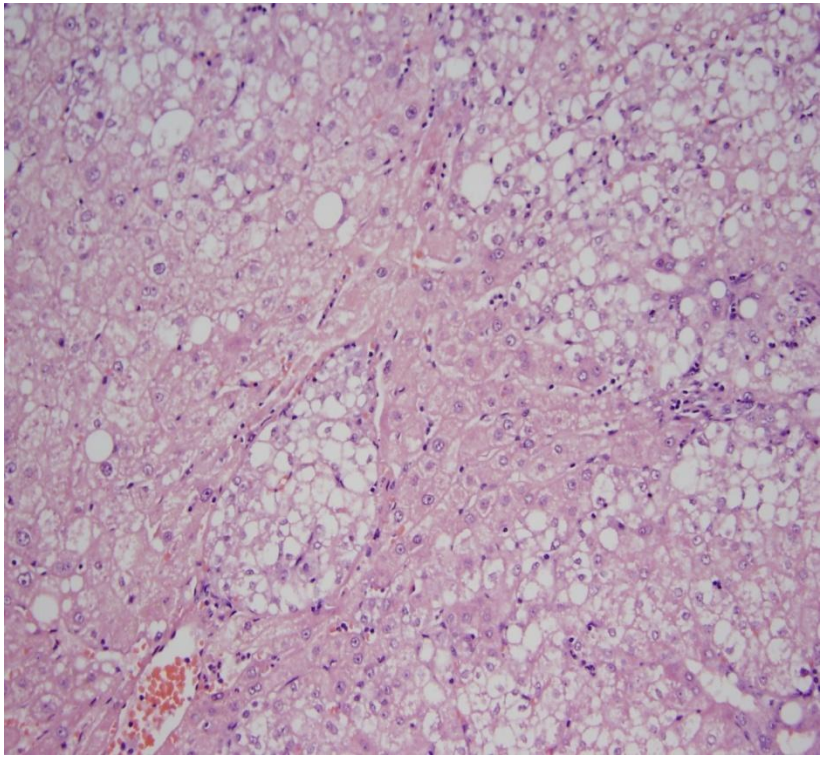
Reticulin Absence

Well-Differentiated HCC: Increased Reticulin with abnormal plates

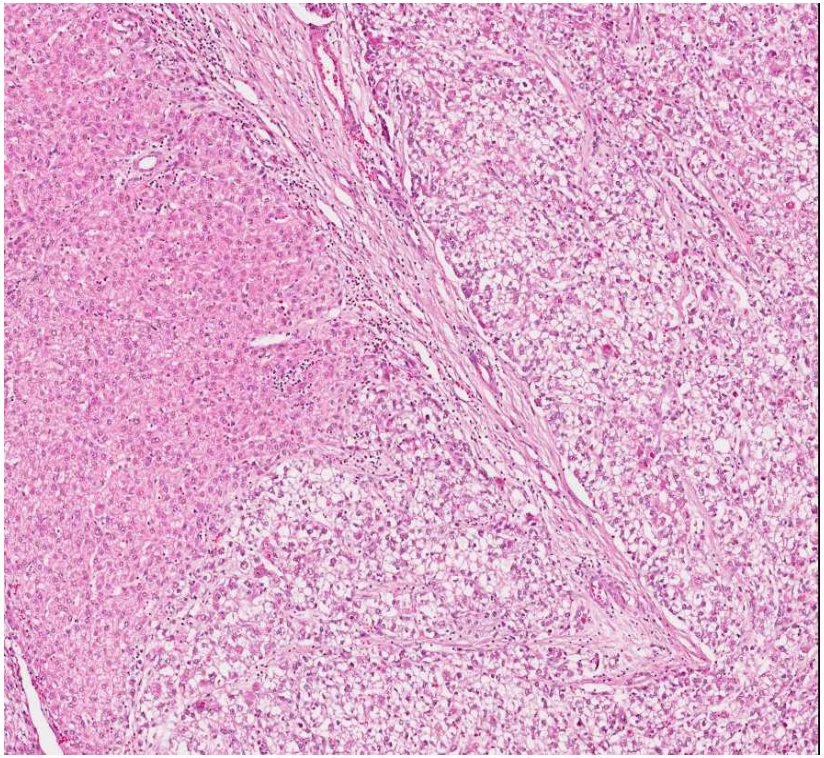


Early HCC: Invasion

Invasion

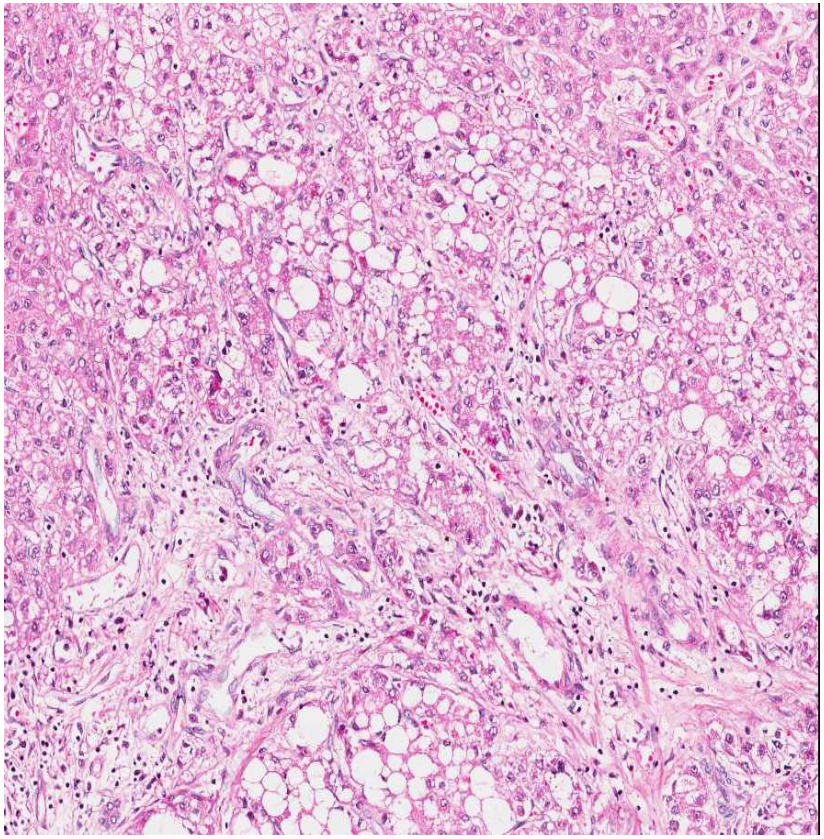


Invasion

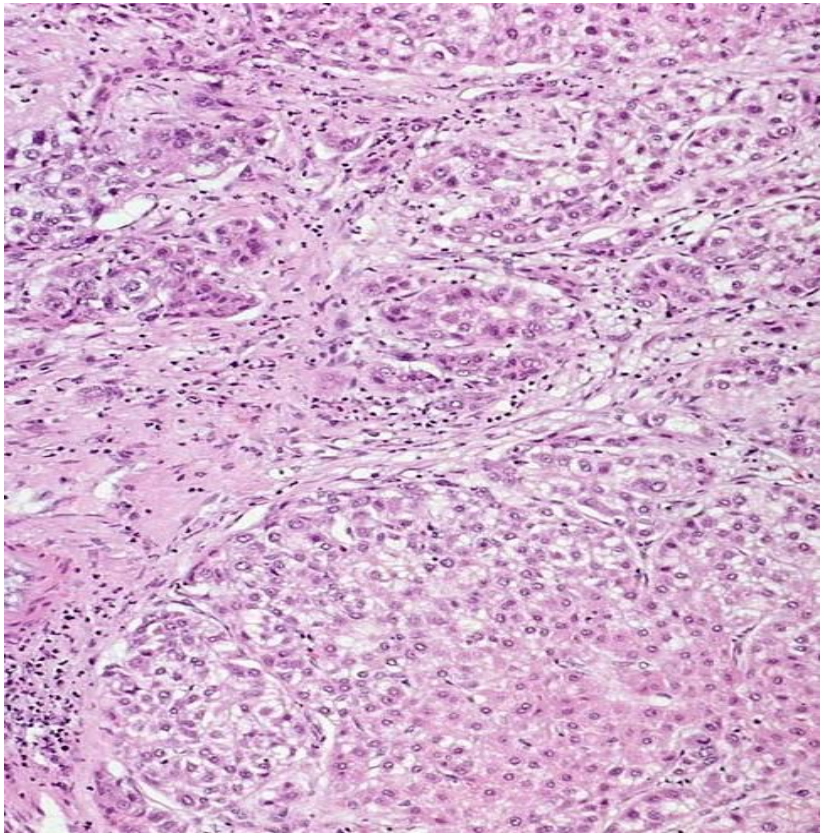


Early HCC: Stromal Invasion

Stromal invasion



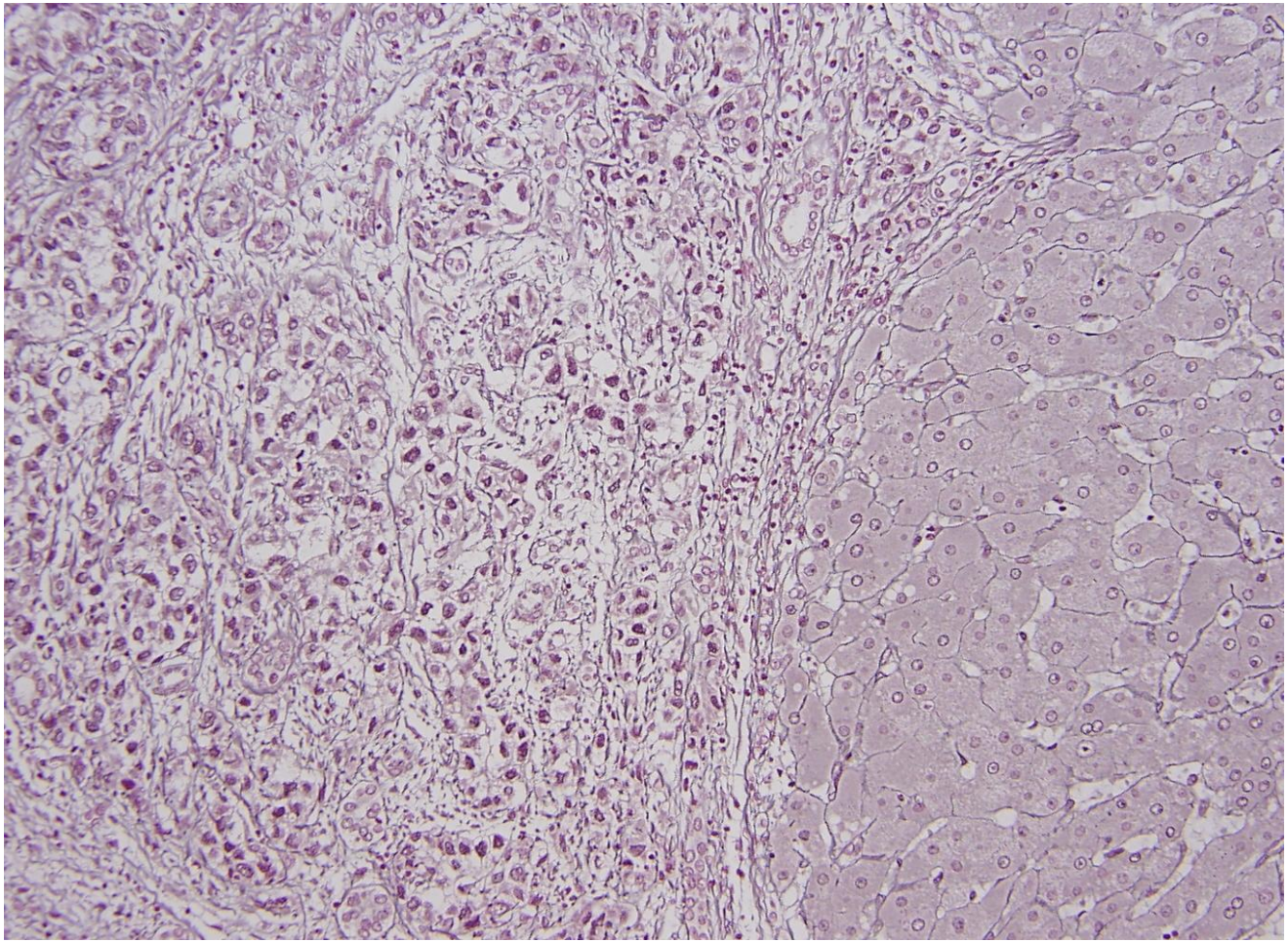
Stromal invasion



Early HCC: Stromal Invasion

RETICULIN

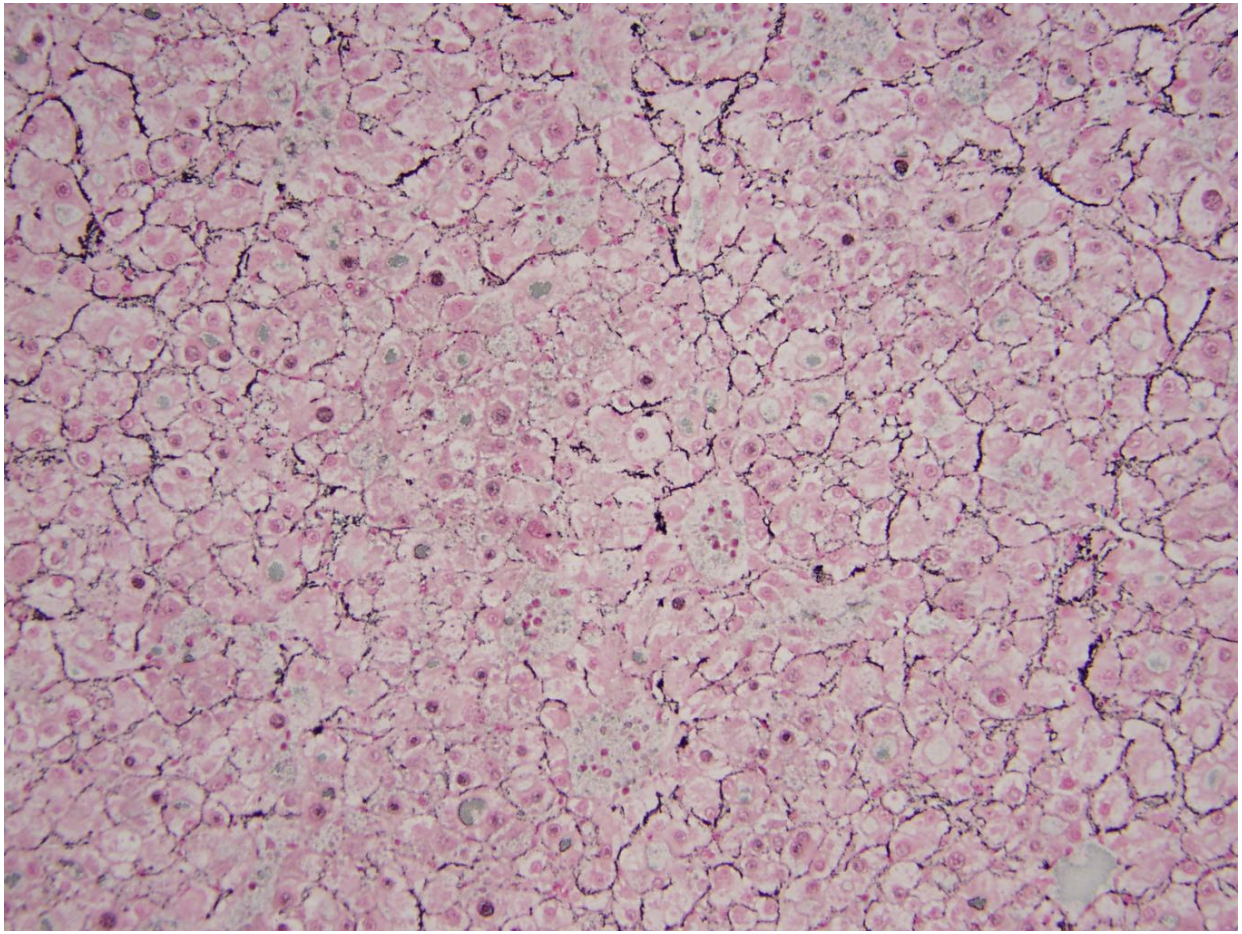
Perinodular stromal invasion



Well-differentiated HCC

RETICULIN

is present but in irregular pattern and amount



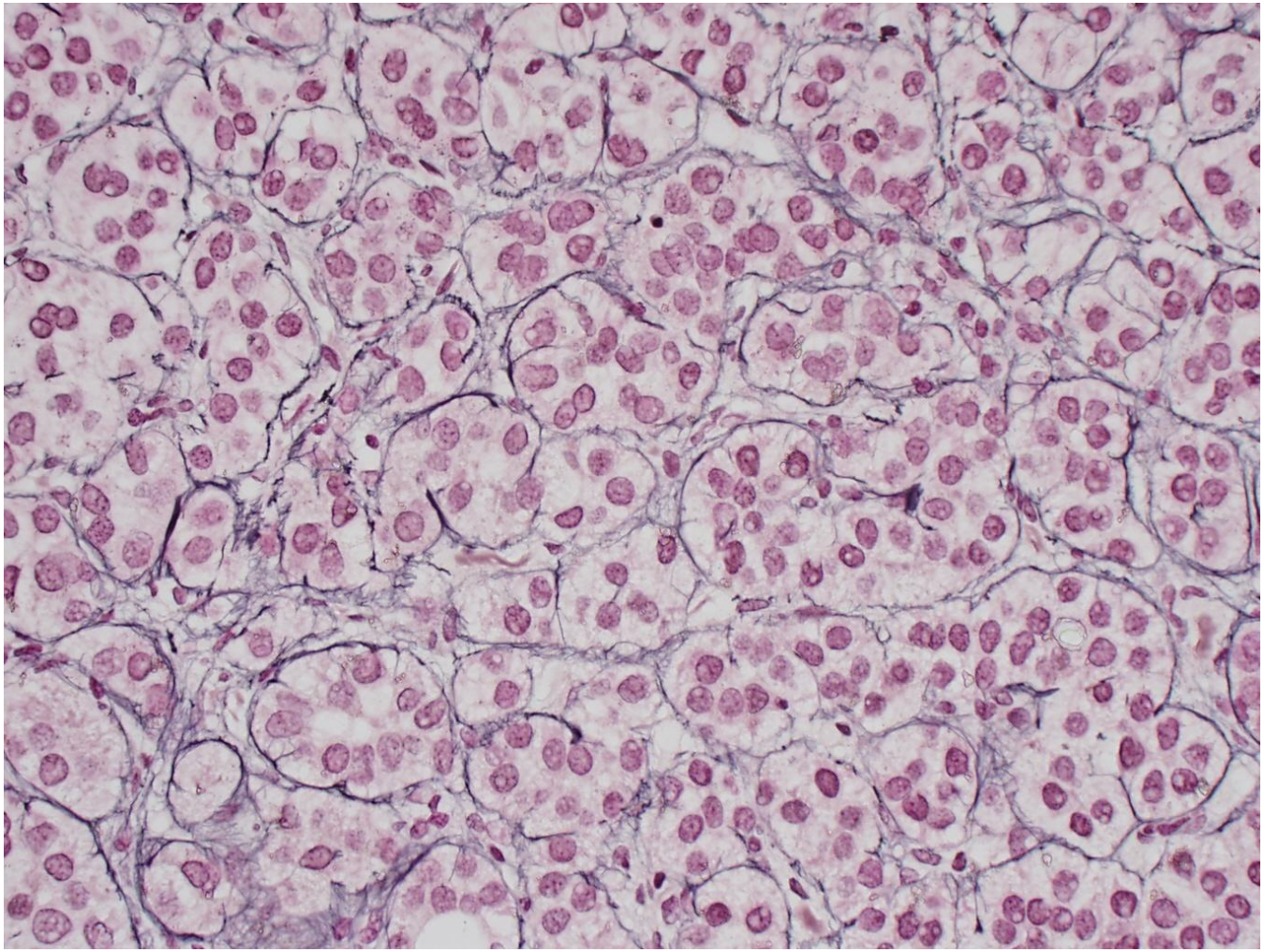
Well-differentiated HCC

RETICULIN

present but in irregular pattern:

Acinar example

Also note small cell change

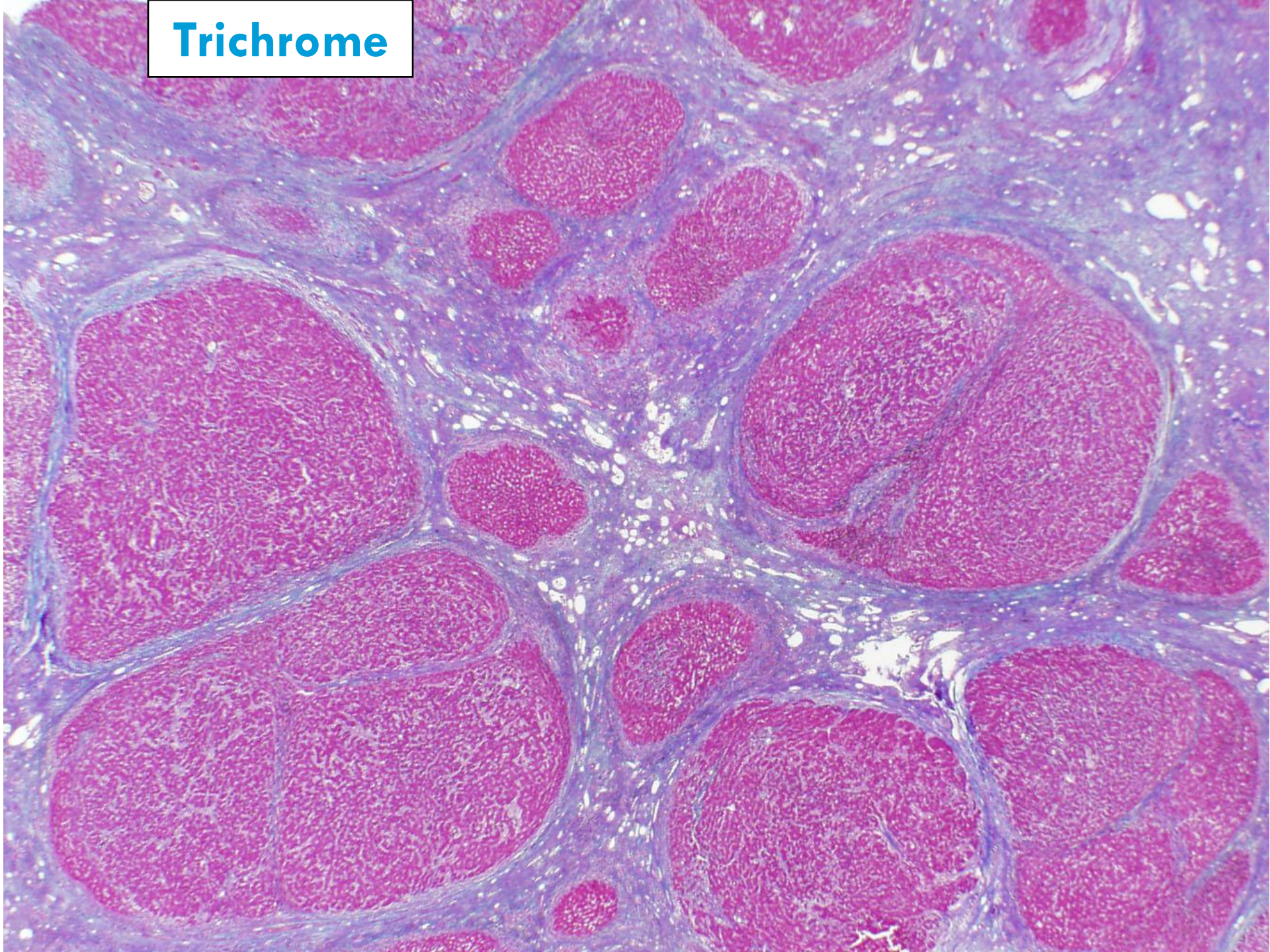


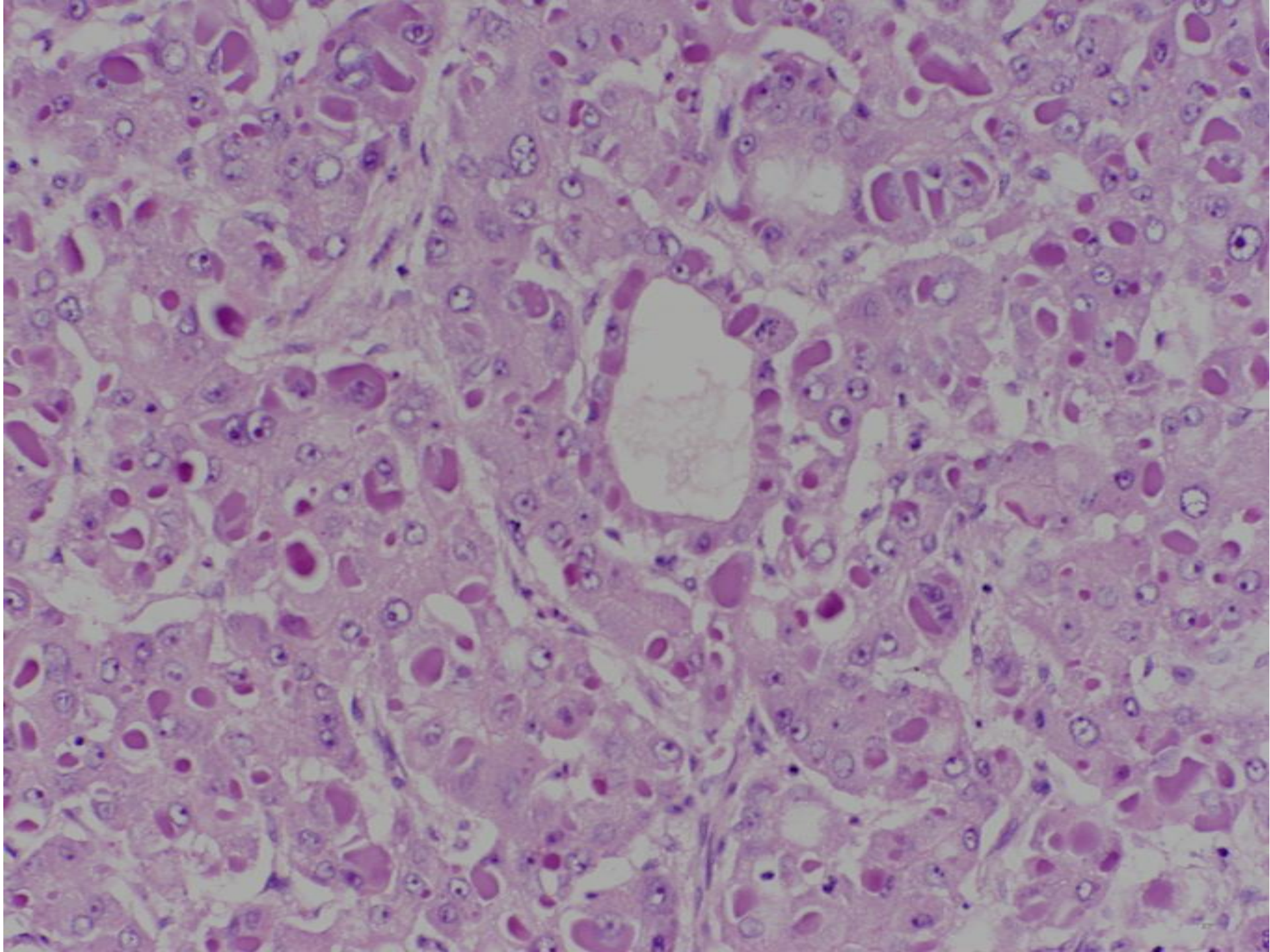
Summary

- **Macroregenerative, Low Grade DN**
  - Minimal cytologic/architectural abnormalities
- **High Grade DN**
  - Moderate cytologic/architectural abnormalities with small/large cell change, decreased reticulin framework
- **Well-differentiated HCC**
  - Small/large cell change
  - Zones of thick plates (>3)
  - Decreased or absent reticulin, or abnormal pattern
  - Early HCC, ≤2 cm: Invasion, stromal and parenchymal

Problem Case: Cirrhotic Liver

- 62-year-old man with end-stage cirrhosis due to HCV
- Followed at UCSF for one year prior to transplant
- AFP not significantly elevated
- No masses noted in liver, other sites
- Sample is from explant


Trichrome




Reticulin

Cirrhosis-like HCC (Variant HCC with Multifocality)

Cirrhosis-like HCC (in cirrhosis)

- Diagnostic problem both clinically and microscopically
- May also have large dominant mass and smaller, cirrhosis like satellite lesions

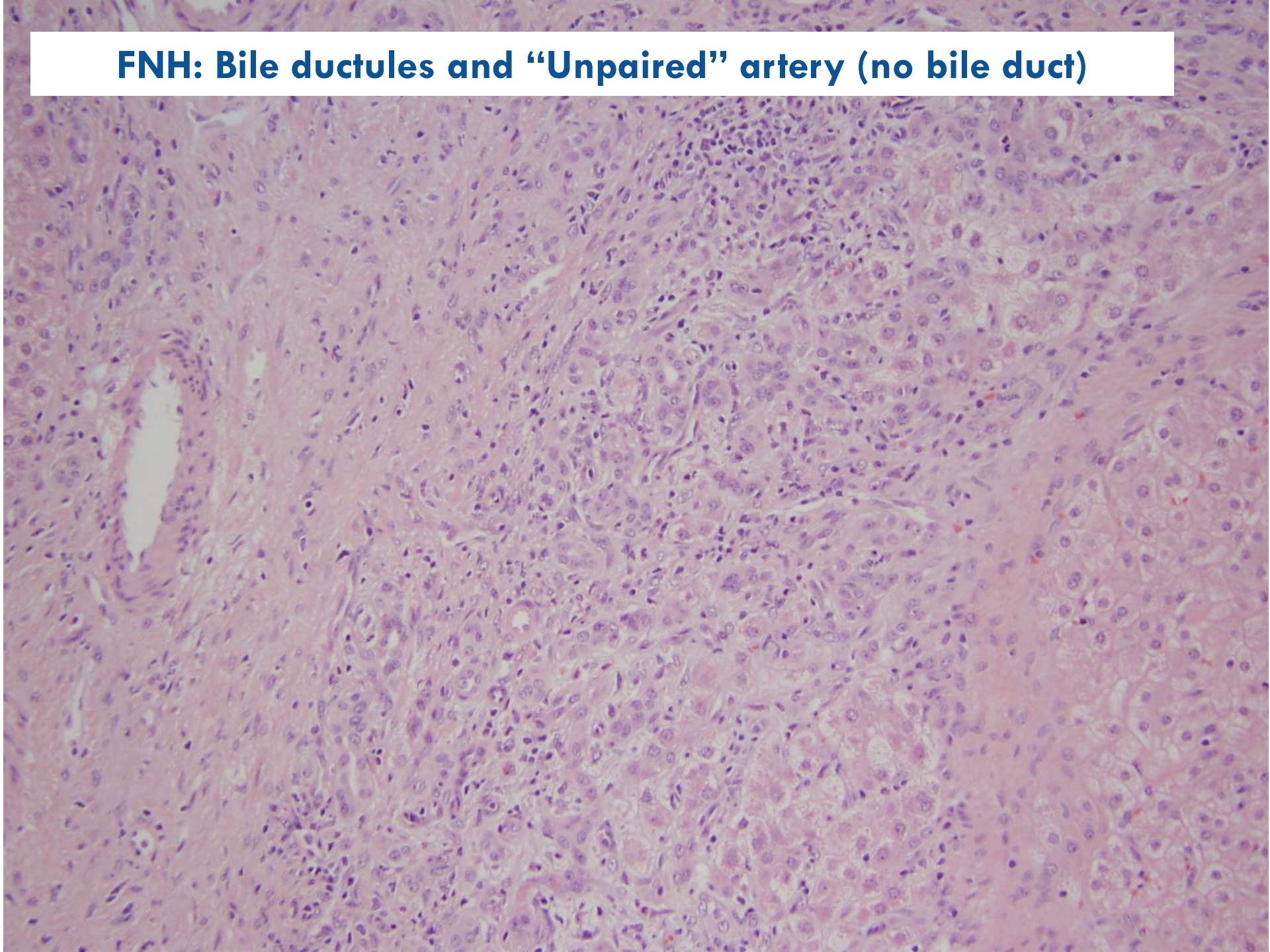
Jakate…Ferrell, et al. Diffuse Cirrhosis-like HCC. AJSP 2010; 34:935-41.

Well-differentiated Hepatocellular Tumors in Noncirrhotic Liver

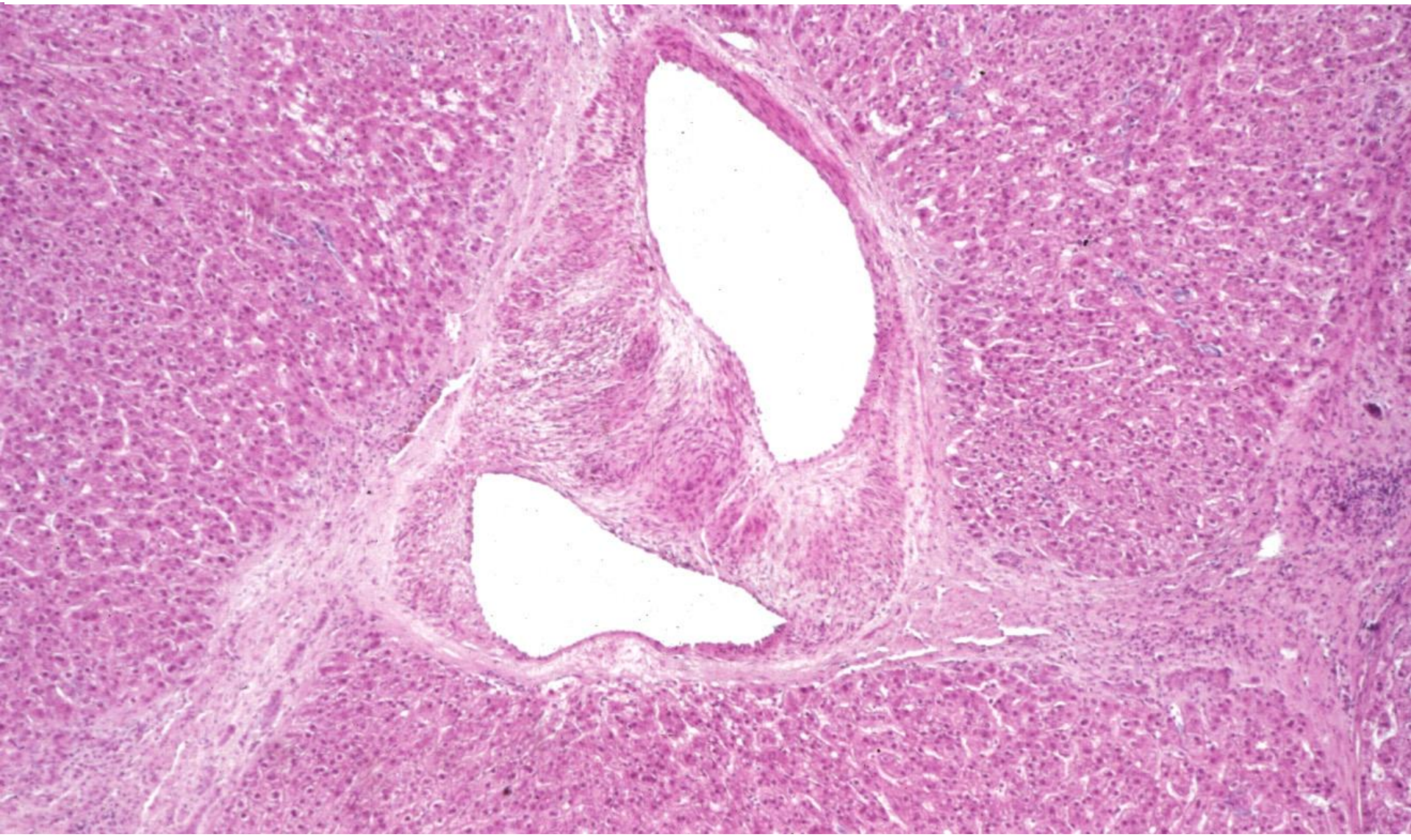
Differential Diagnosis

- Focal nodular hyperplasia (FNH)
- Hepatocellular Adenoma (HCA)
- Hepatocellular Carcinoma (HCC)

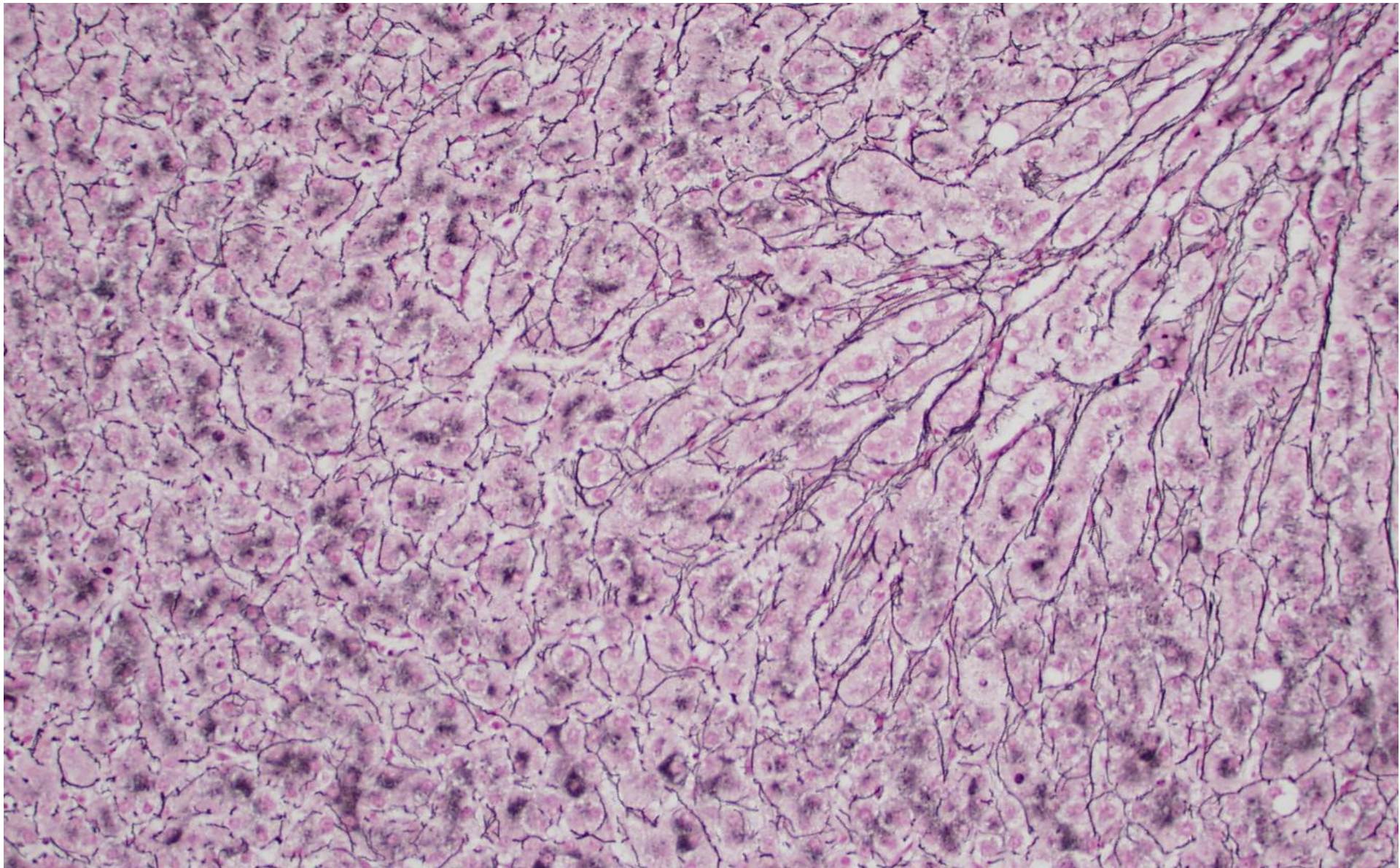
FNH: Bile ductules and “Unpaired” artery (no bile duct)



Another FNH: Note abnormal "dystrophic" thick walled vessels, often similar to those seen in AV malformation surrounded by connective tissue



FNH, Reticulin stain: abundant framework present, but plate width and shape can be variable in rare cases, so can overlap some variants seen in HCC.



FNH – Core Biopsy

- **Don't mistake this lesion for:**
  - *Scar zones with arteries and lymphoid infiltrates of ductopenic portal areas*
    - **Hepatocytes at edge of scar zones can be positive for copper in both FNH and chronic obstructive biliary disease**
  - *Reaction to adjacent lesion*
  - *Thicker plates of HCC*

Adenoma (HCA):
New Classifications, WHO 2010

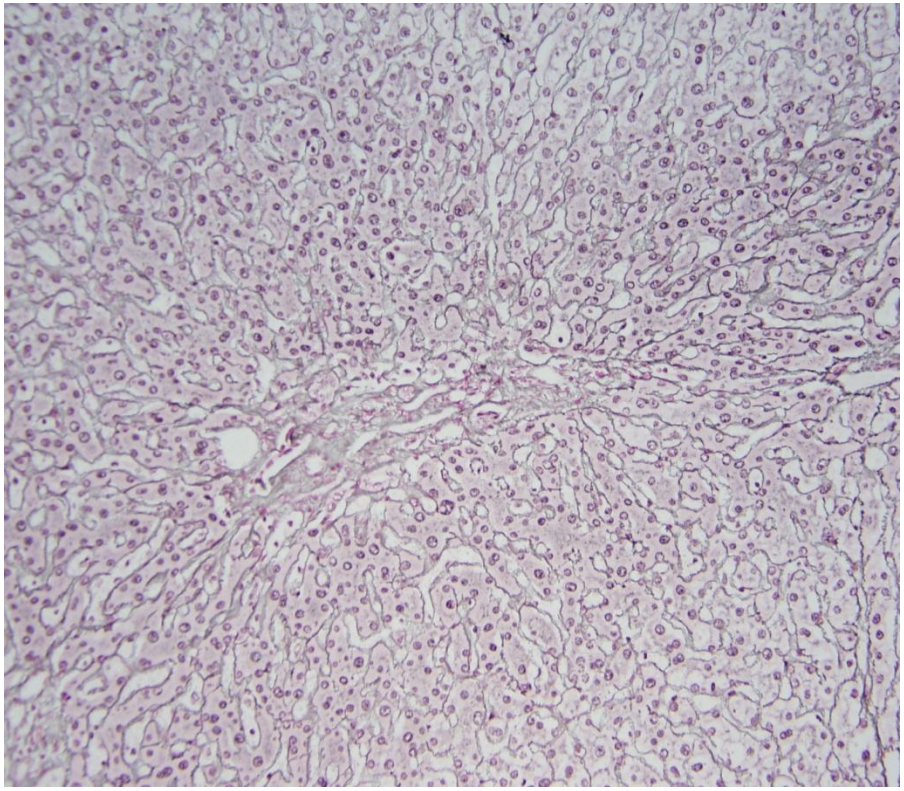
- **Adenoma: 4 variants**
  - **Variant 1. HNF1α mutations, 40-50%**
    - **Fatty change, no atypia, no inflammatory infiltrates**
  - **Variant 2. β-catenin mutation, <10%**
    - **very high risk for HCC**
  - **Variant 3. Inflammatory adenoma**
    - **formerly known as telangiectatic FNH**
  - **Others (Variant 4?)**
    - **no specific trait**

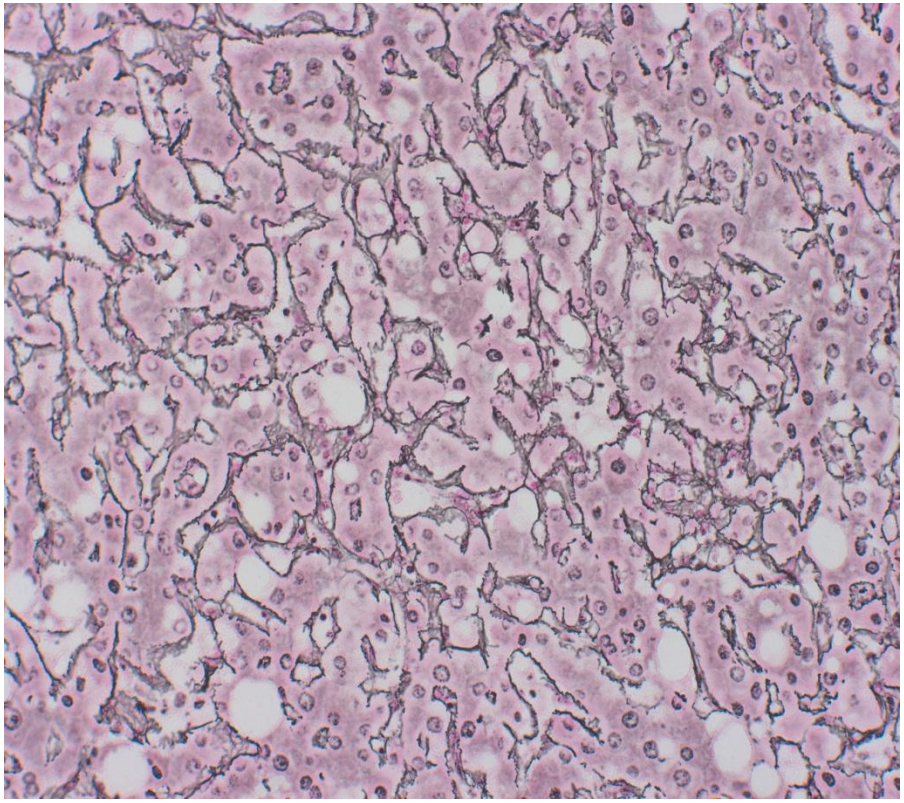
Hepatocellular Adenoma: Common Features

- Association with estrogen such as OCP (obesity??), diabetes
- Risk of hemorrhage correlates with increase in size (usually >5 cm)
- Rare risk of HCC
- Histology: bland cytology, intact reticulin framework, no mitoses, no ductules

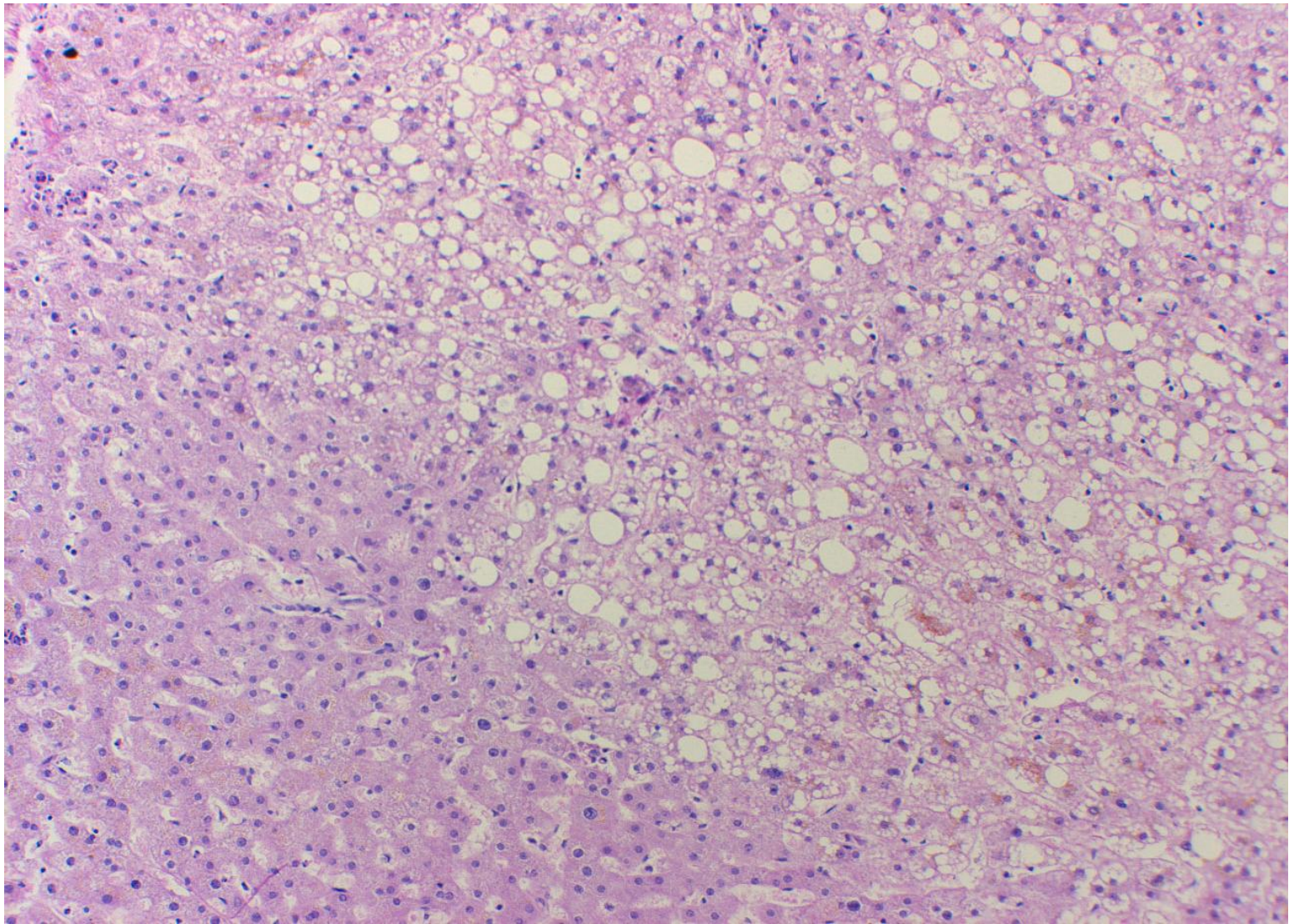
Hepatocellular Adenoma: Common Features

Reticulin framework intact, plate architecture and cytology mimics normal patterns





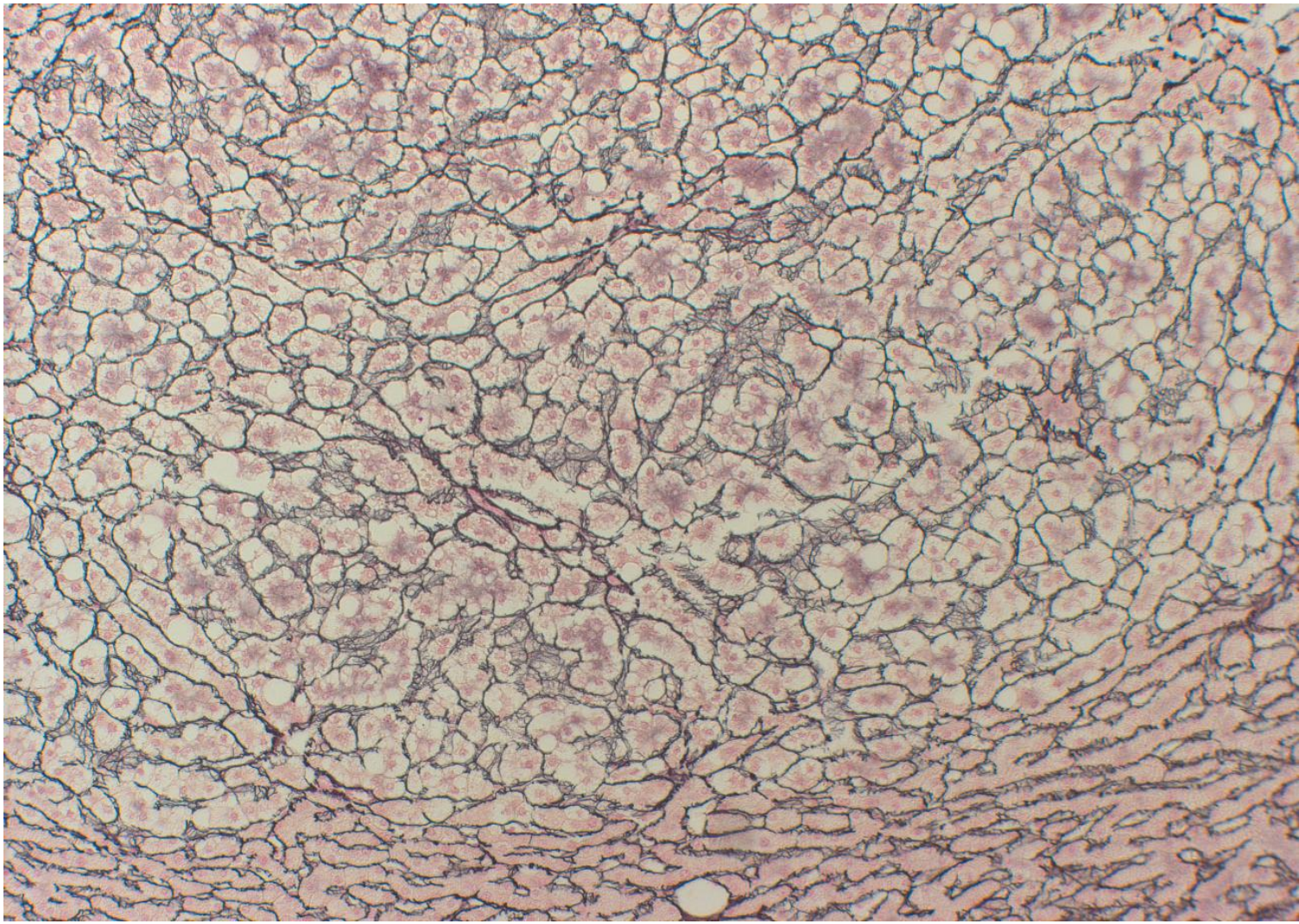
Variant 1 Hepatocellular adenoma, HNF1α type



Variant 1 Hepatocellular adenoma, HNF1α type

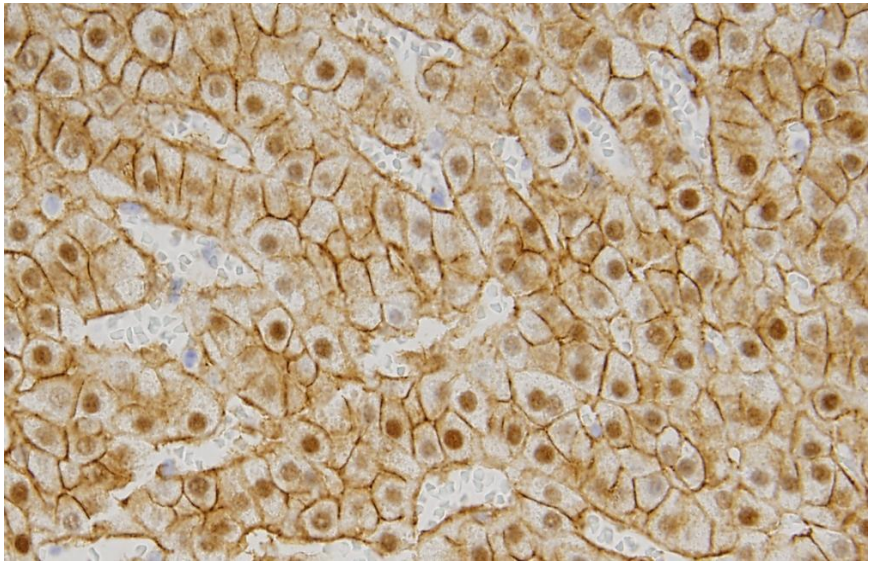
Reticulin

Small acini

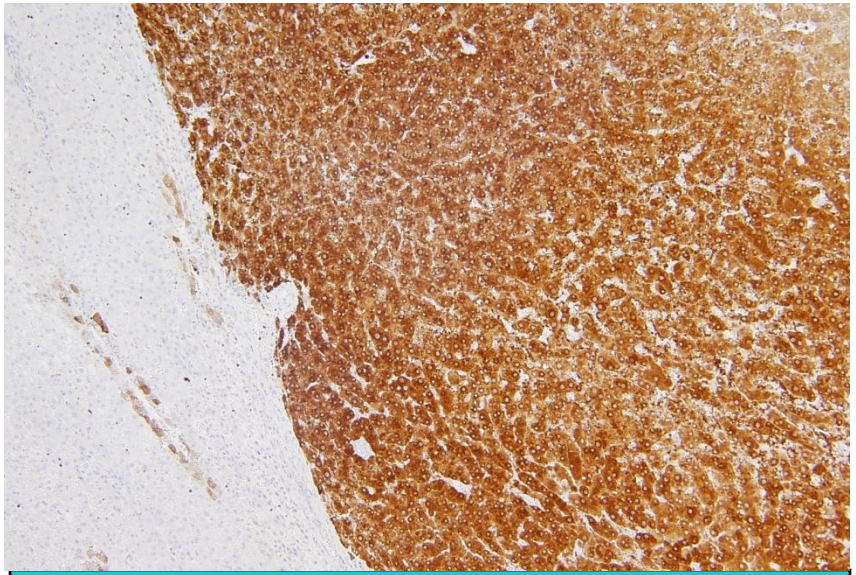


Variant 2 Hepatocellular adenoma: B-catenin-mutated type

Rare adenomas with HIGH risk of HCC transformation
Association with male hormone use



B-catenin nuclear staining

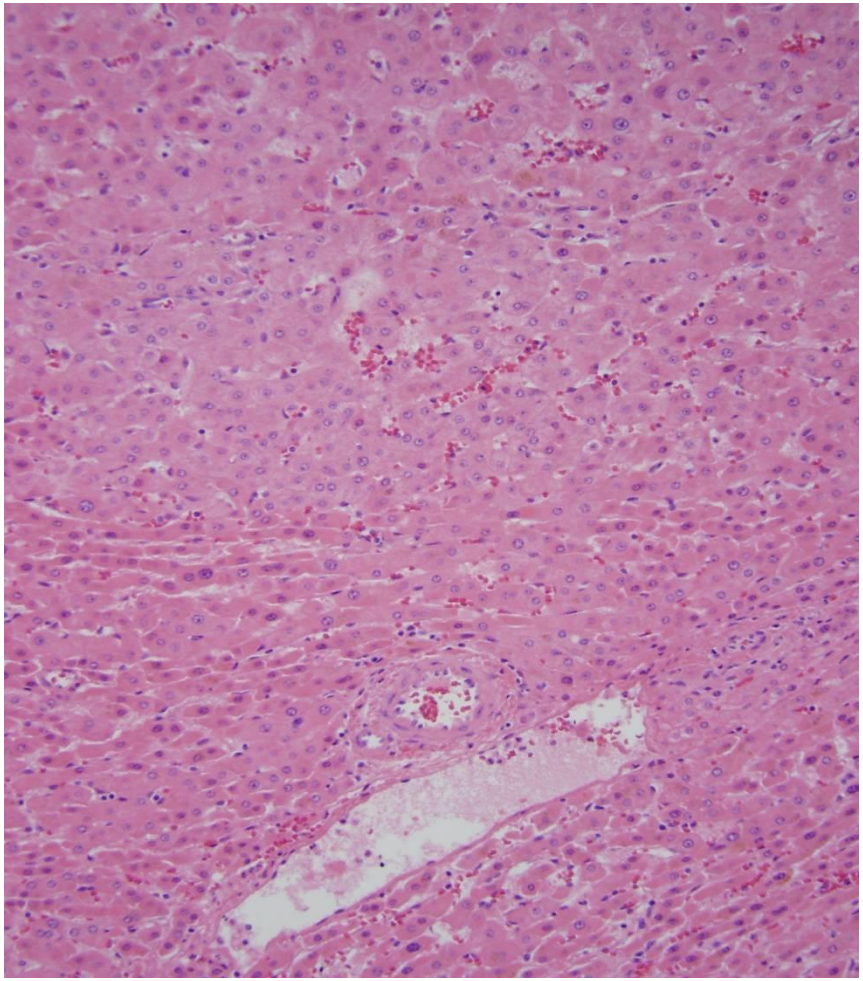


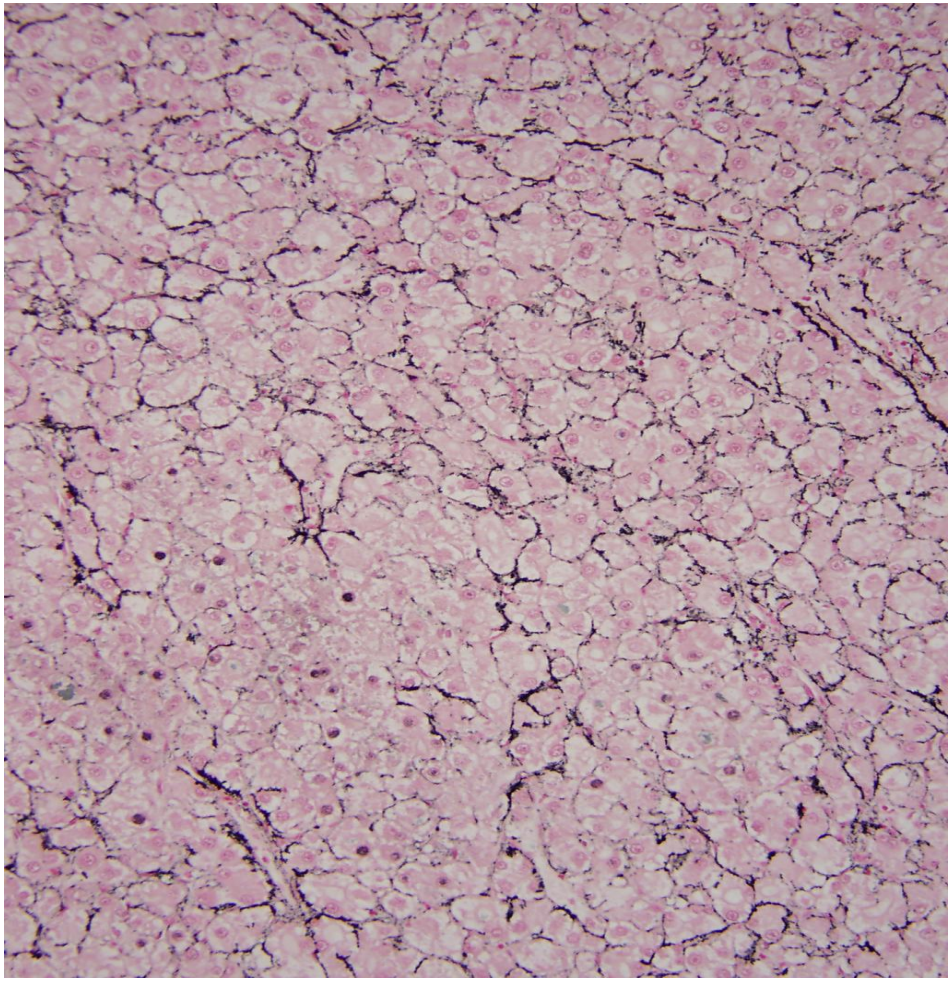
Glutamine synthetase: Intense, diffuse staining

Bioulac-Sage et al. HCA subtype classification using molecular markers and IHC: Hepatol 2007;46:740-8.

Variant 2 Hepatocellular adenoma: B-catenin-mutated type: High risk for HCC

Adenomas with HCC features: Loss of reticulin





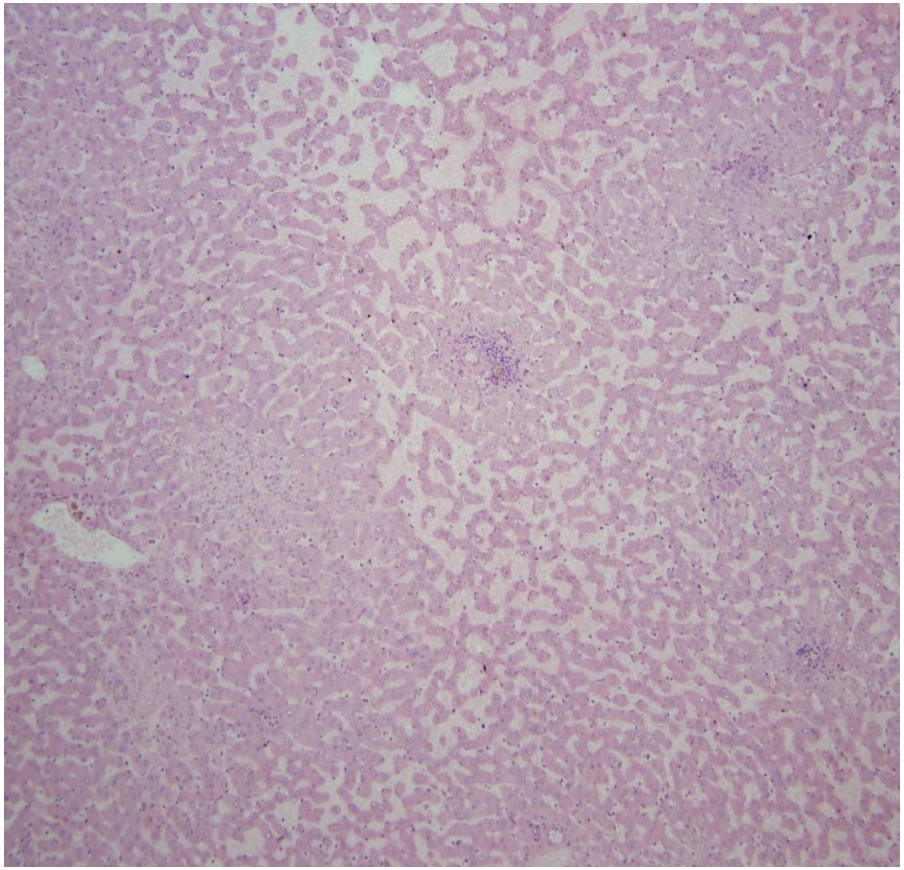
Variant 3 Hepatocellular adenoma, inflammatory type

- *Formerly known as Telangiectatic FNH*
- Features include:
  - Dilated sinusoids
  - Focal inflammatory change around arteries
  - Arteries can be in clusters
  - Mild ductular reaction

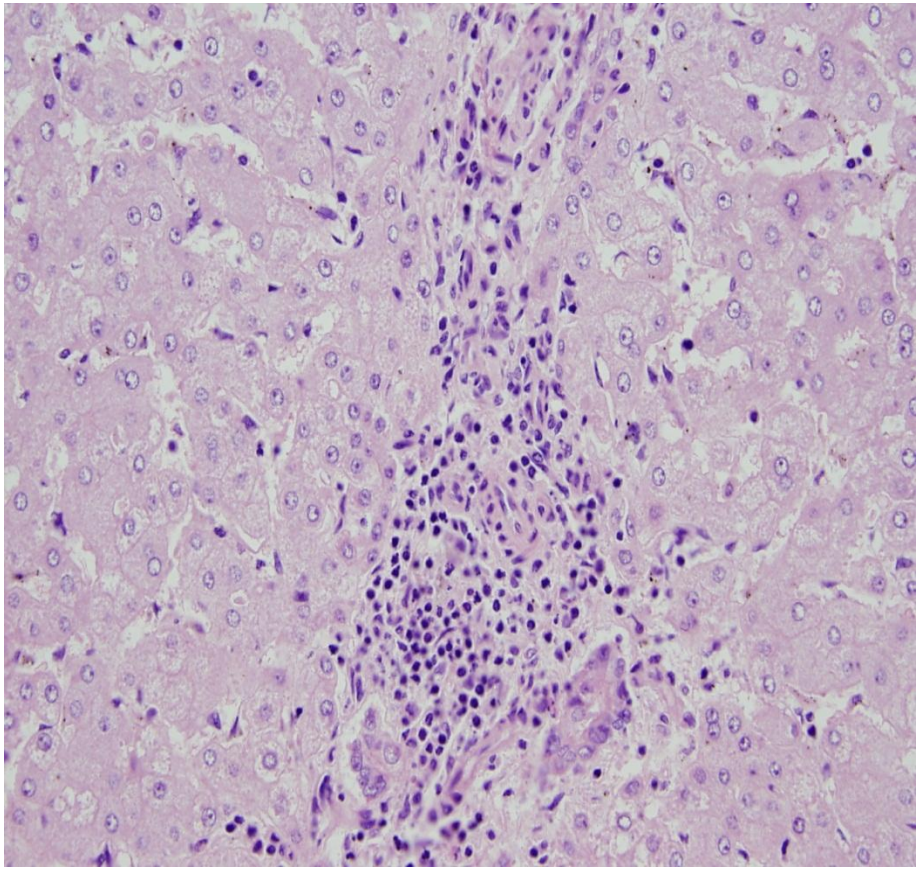
  - *Association with obesity*
  - *Also seen in males*

Variant 3 Hepatocellular adenoma, inflammatory type

Dilated sinusoids, focal inflammation around arteries



Ductules with artery and inflammation




Ductule present in this zone with arteries;
unremarkable hepatocytic cytology.

Adenomatosis

- Large numbers of adenomas (>10)
- May be most common in:
  - HNF1α, type 1 variant
  - Inflammatory variant, type 3

FNH and HCA

- Usually young patient, noncirrhotic liver
- Key features:
  - Abnormal vessels, small or large
  - Intact reticulin framework
- Problem features:
  - Architectural irregularities with thicker plates or acinar change
  - Cytologic atypia (more common in FNH)
  - Sampling problems – don't get diagnostic areas
  - Both FNH and HCA, inflammatory type with ductular reaction

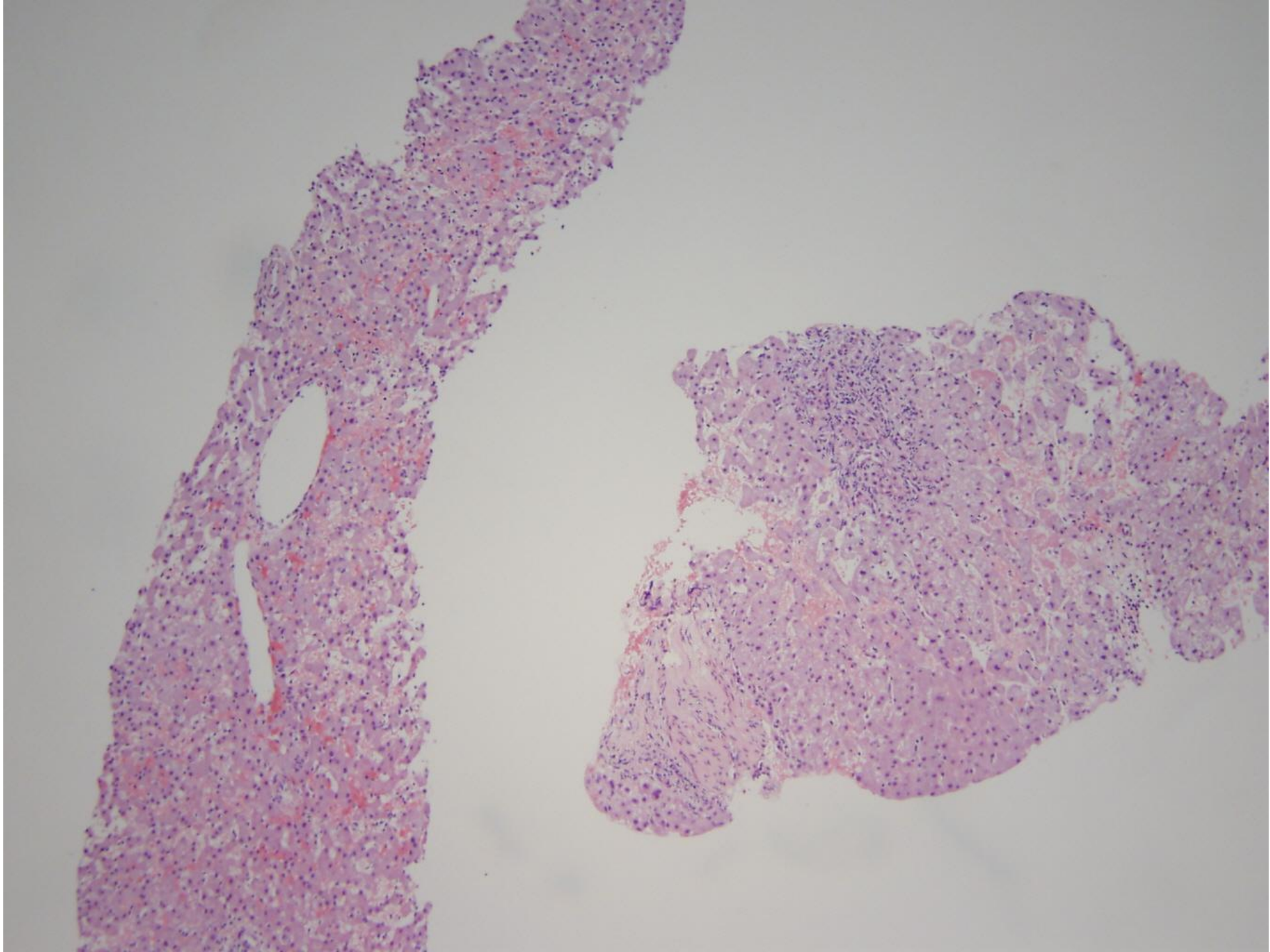
Problem Case Example

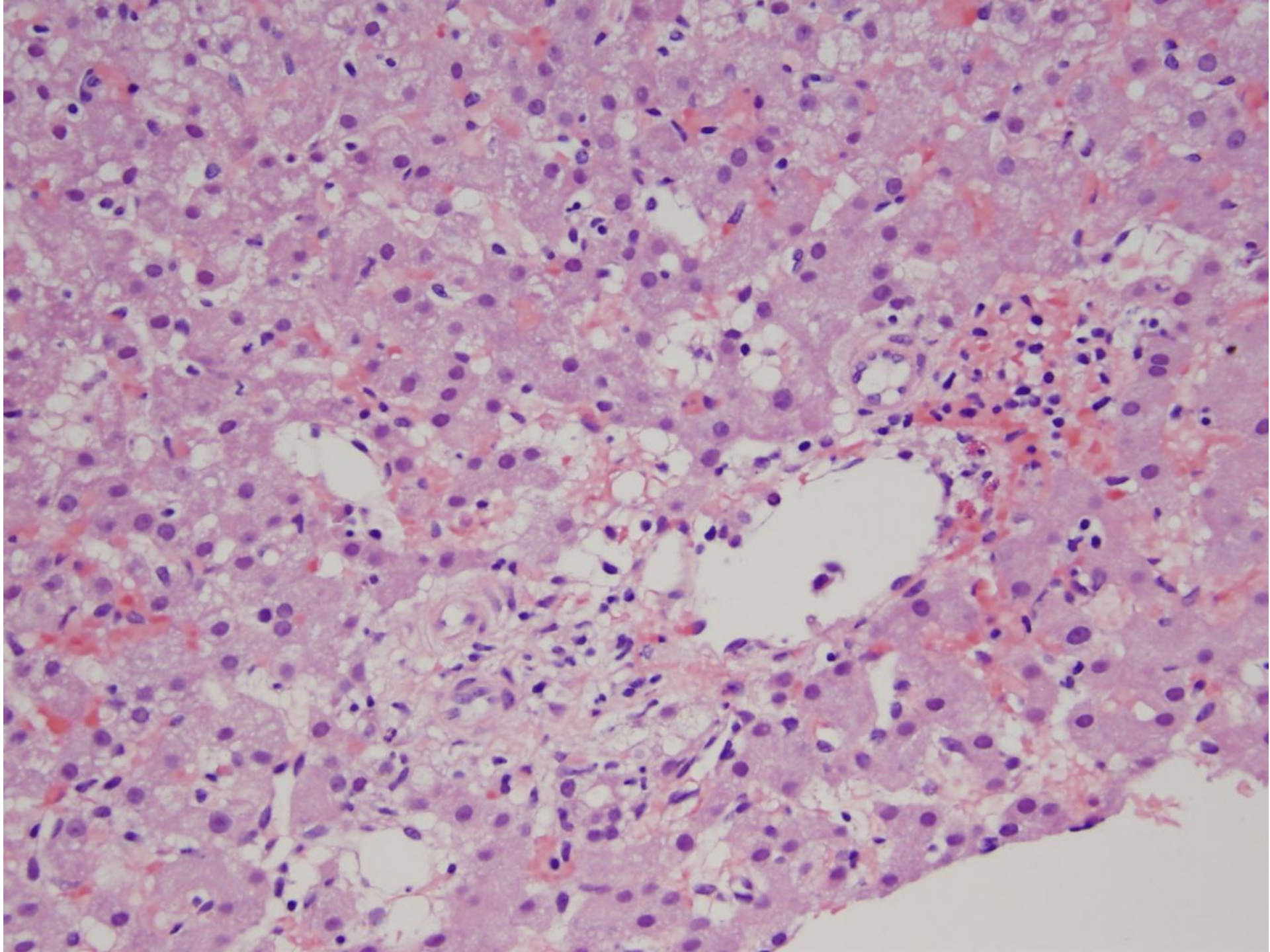
To Illustrate use of new immunohistochemistry tools in Differentiation of FNH and hepatocellular adenoma (HCA)

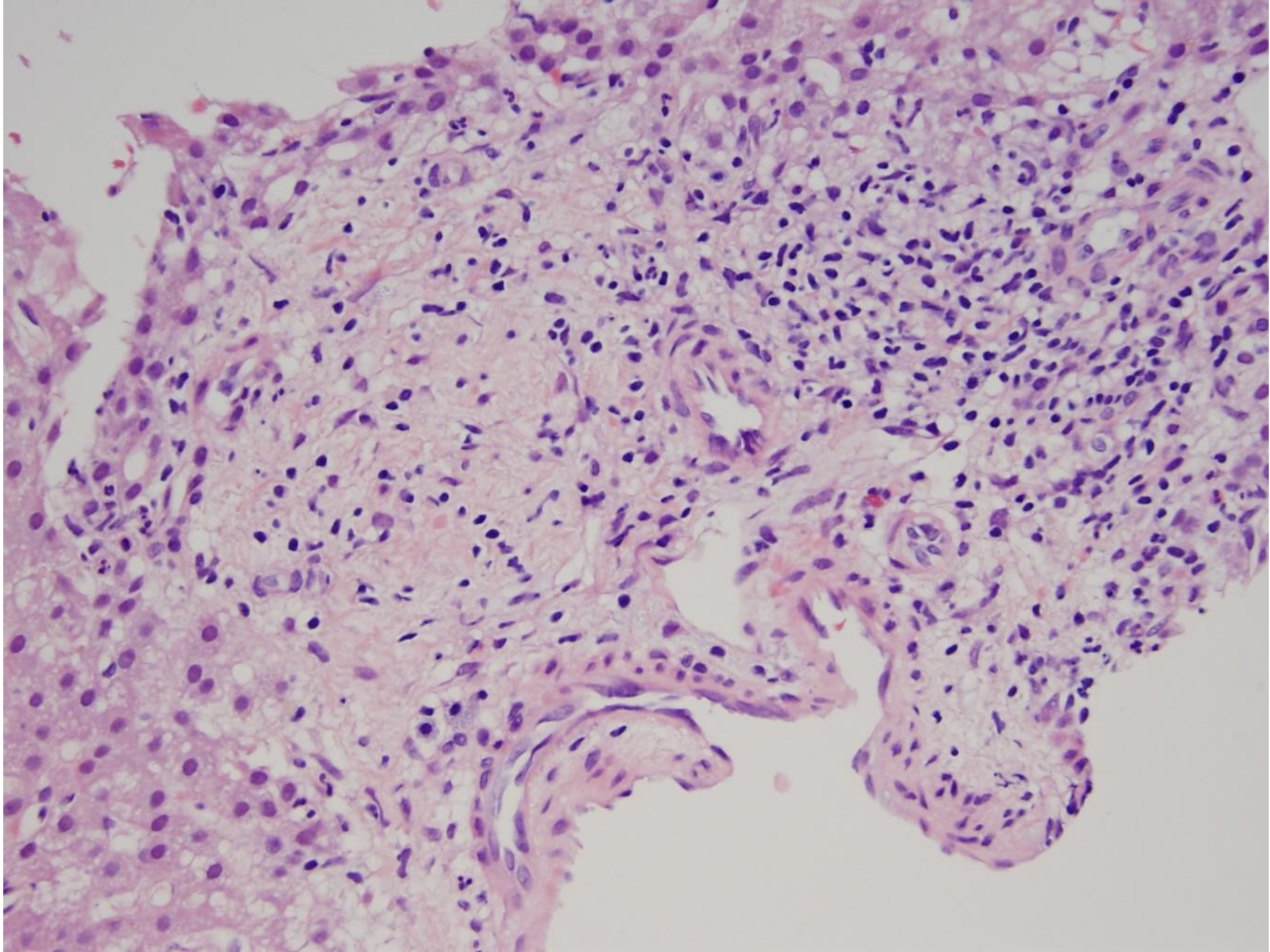
Case History (Question: FNH or HCA?)

- 48-year-old woman
- Multiple liver masses, largest 13 cm and 4 cm
- Presented with abdominal pain and elevated alkaline phosphatase
- No history of liver disease or other tumors
- Imaging differential diagnosis:
  - Hepatocellular carcinoma (well differentiated or fibrolamellar type)
  - Focal nodular hyperplasia
  - Adenoma/adenomatosis
  - Hemangioma
  - Metastatic lesion

NEEDLE BIOPSY PERFORMED







Immunostains: FNH vs. HCA

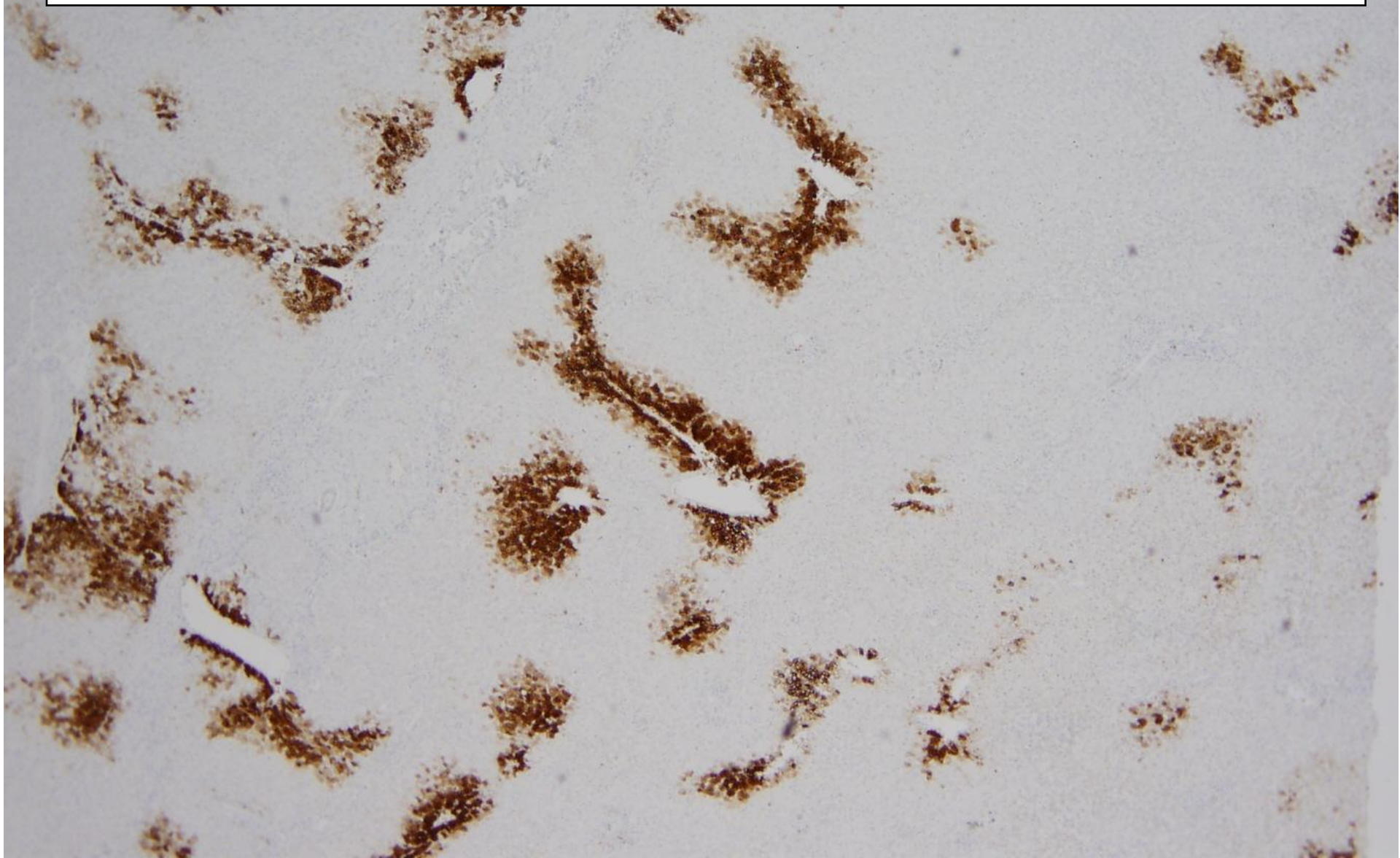
THREE STAINS

- Glutamine synthetase (GS)
- Keratin 7(K7)
- Amyloid A (SAA)

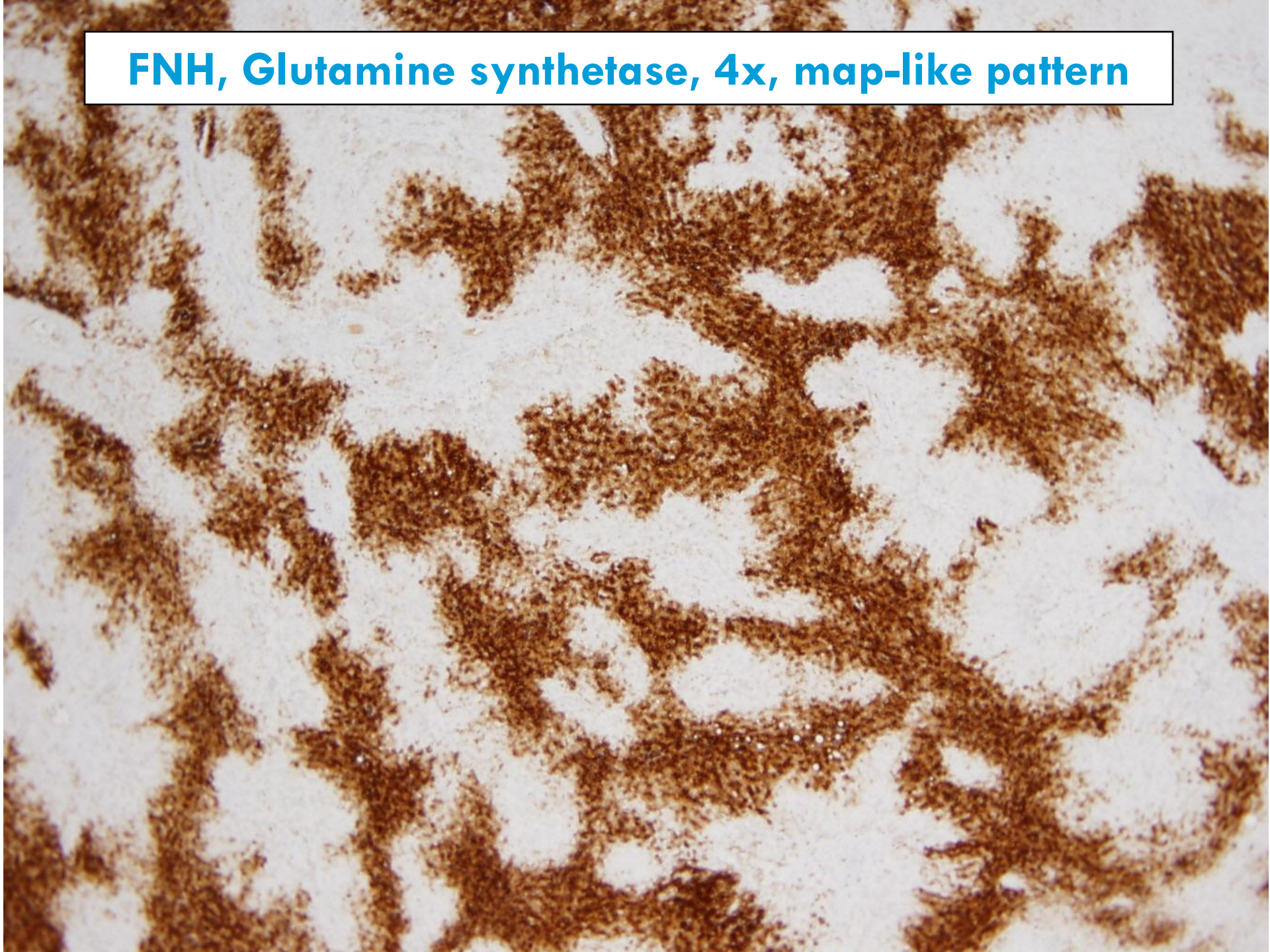
Immunostains: FNH vs. HCA

- **Glutamine synthetase (GS)**
  - **Enzyme typically in centrizonal hepatocytes in normal liver**
  - **More extensive "map-like" pattern in FNH**
  - **Variable pattern in other settings but typically located around veins in HCA**
- **Keratin 7(K7)**
- **Amyloid A (SAA)**

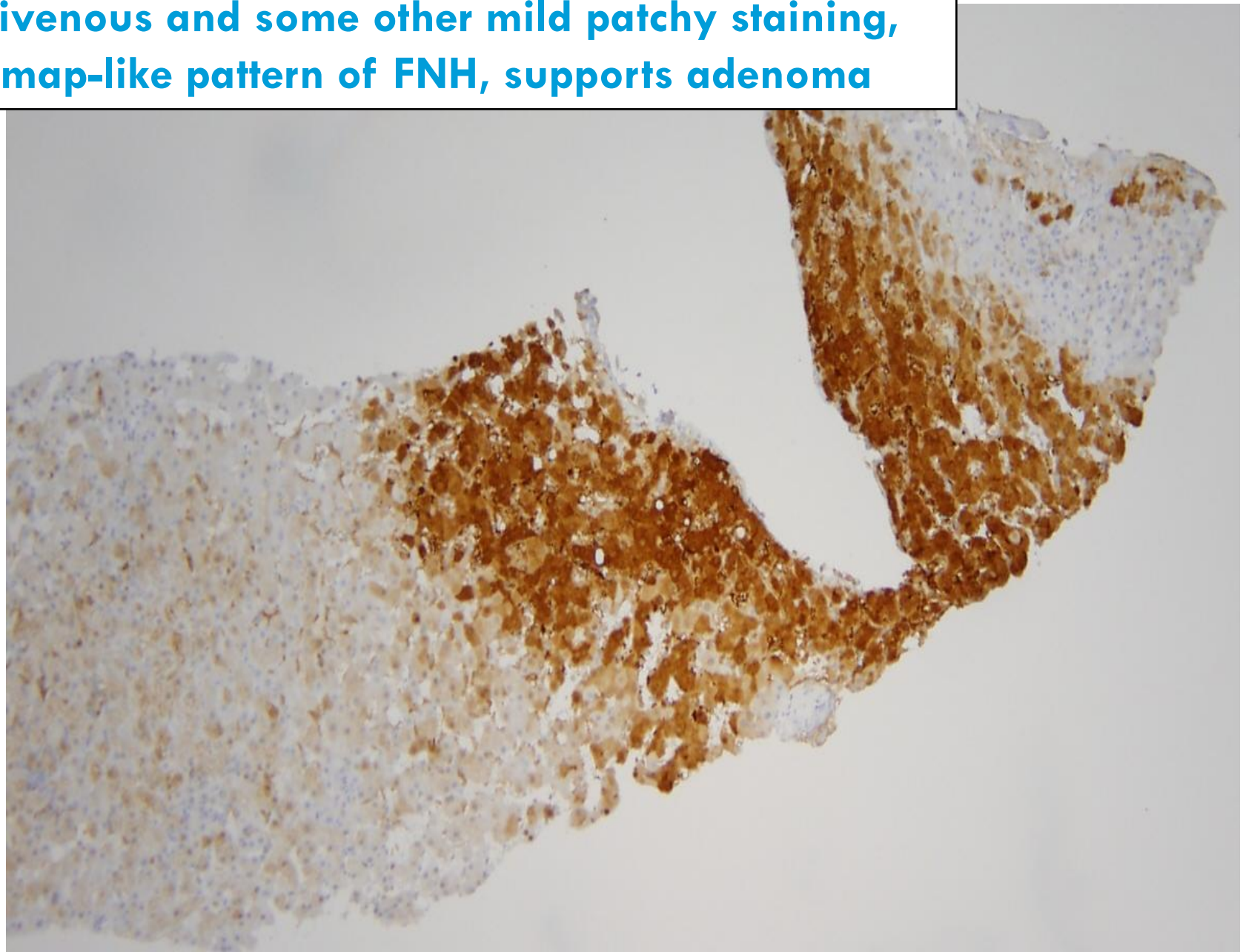
Glutamine synthetase, 10x:
Normal pattern: perivenous staining, normal liver and HCA



FNH, Glutamine synthetase, 4x, map-like pattern



Our Case Example: Glutamine synthetase, 10x:
Perivenous and some other mild patchy staining, no map-like pattern of FNH, supports adenoma

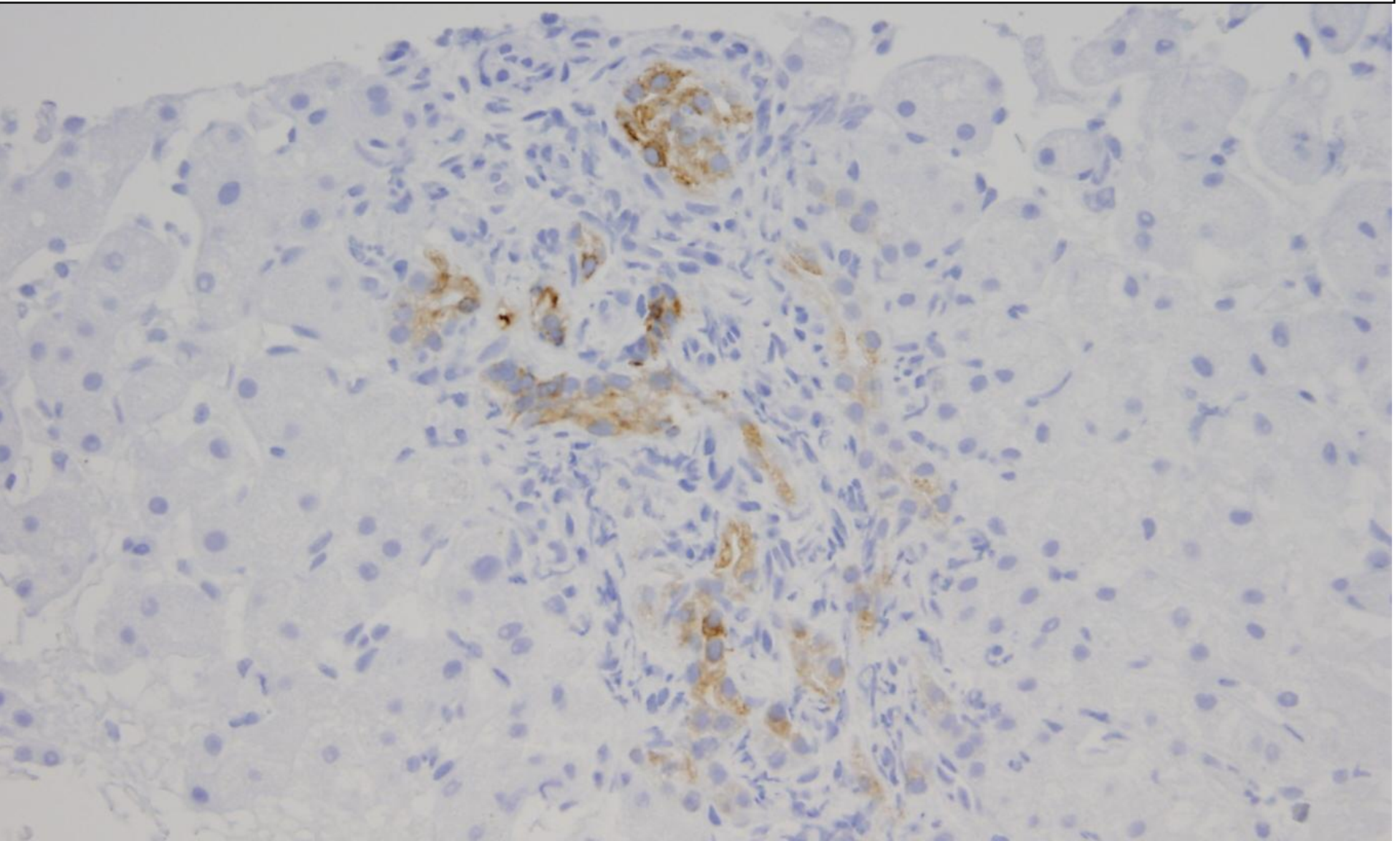


Immunostains: FNH vs. HCA

- Glutamine synthetase
- **Keratin 7 (K7)**
  - **Highlights ductular reaction**
- Amyloid A (SAA)

Case, K7, 40x: Focal ductular reaction in arterialized zones, consistent with inflammatory HCA

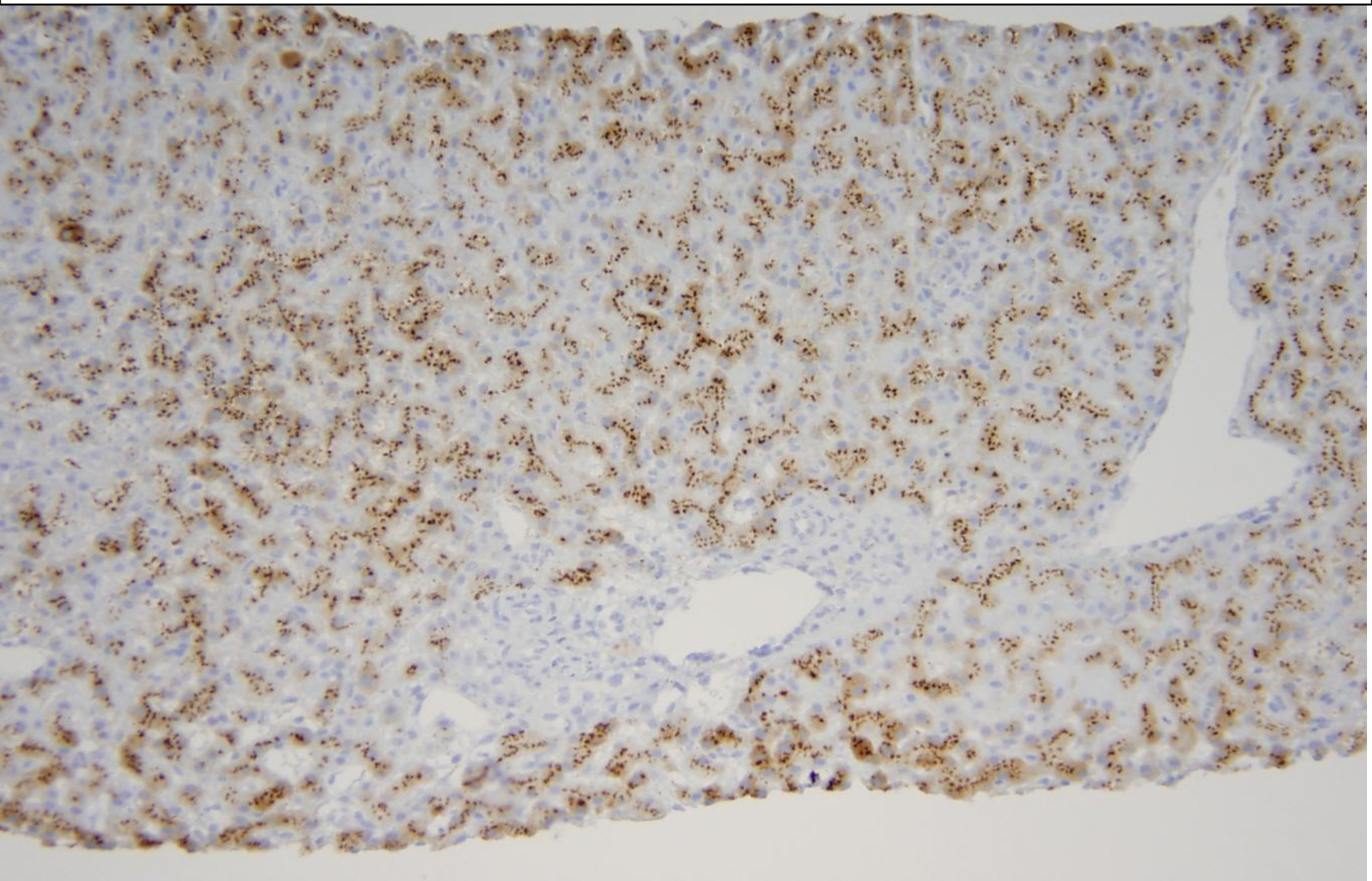
Note: staining may be intermediate in intensity



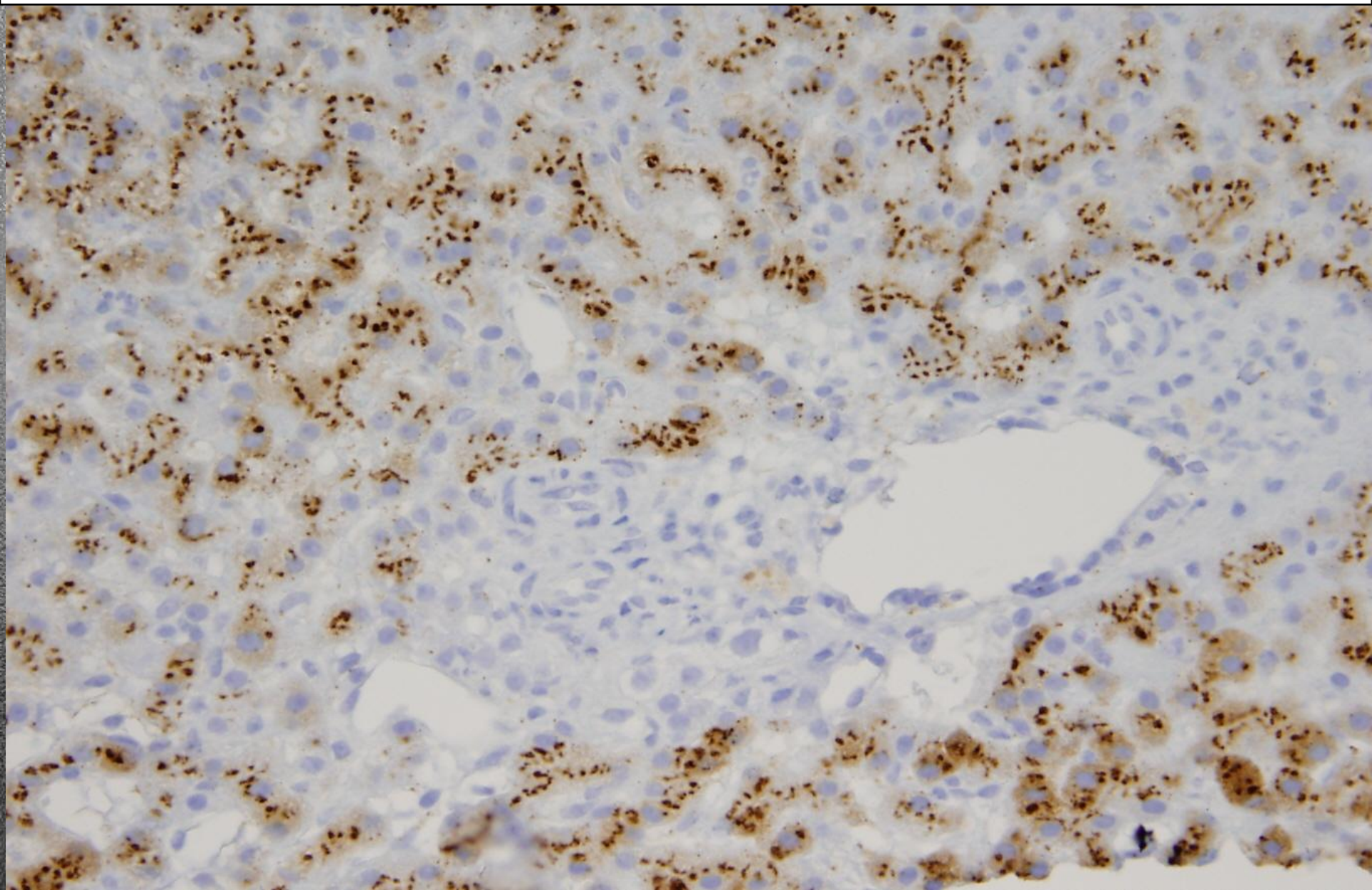
Immunostains: FNH vs. HCA

- Glutamine synthetase
- K7
- **Amyloid A (SAA):**
  - **Reactive/inflammatory form of amyloid**
  - **Stains as granular deposits in hepatocytes**
  - **Prominent staining in Inflammatory HCA**
  - **Can be seen in background liver**
  - **Limited staining in FNH**

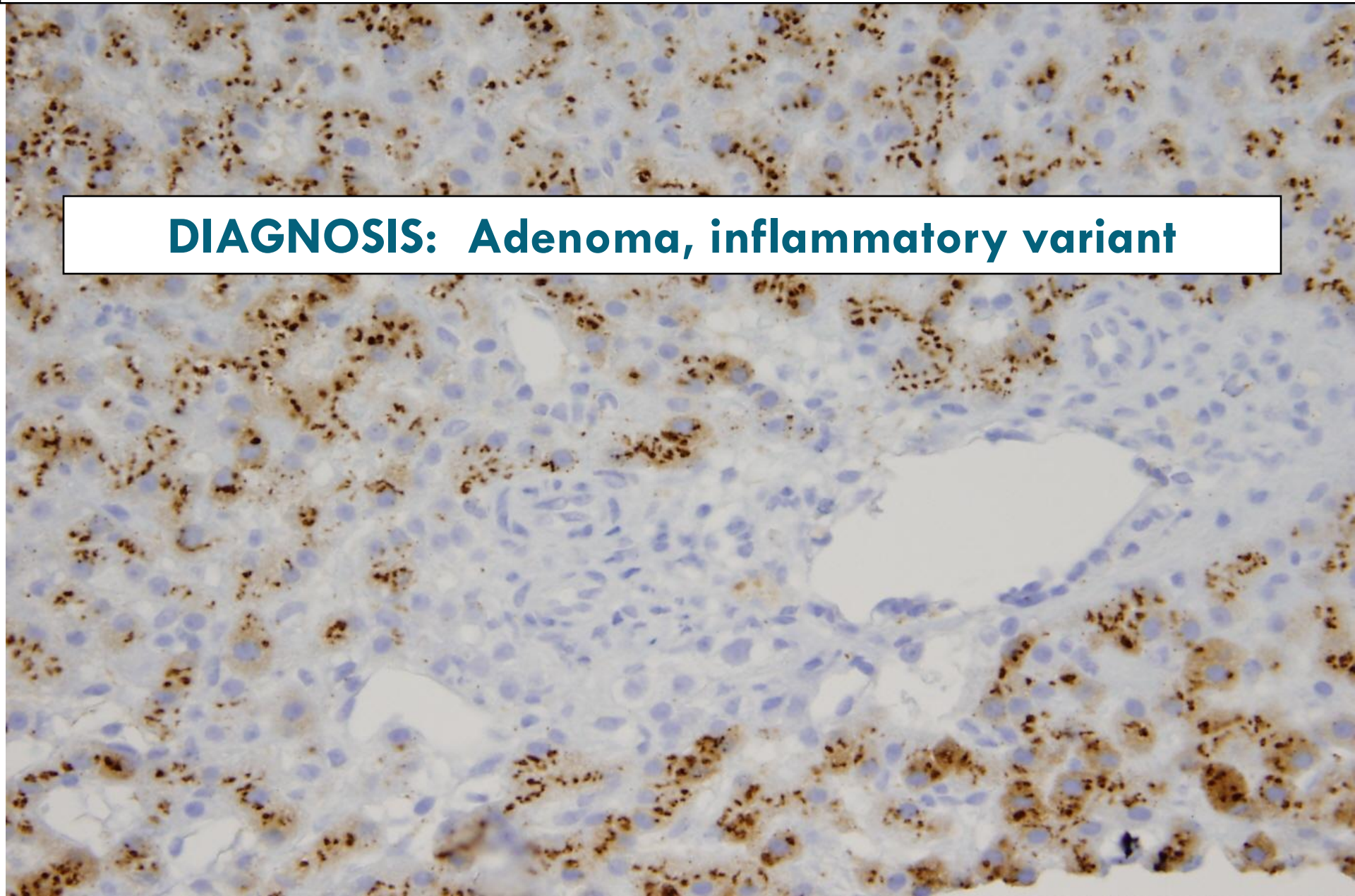
Case, Amyloid A (SAA), 20x: diffuse cytoplasmic, granular staining of hepatocytes



Case, Amyloid A (SAA), 40x: diffuse cytoplasmic, granular staining of hepatocytes



Case, Amyloid A (SAA), 40x: diffuse cytoplasmic, granular staining of hepatocytes



Immunostains: FNH vs. HCA

- **Glutamine synthetase**
  - **Map-like, extensive dense staining in FNH**
- **K7**
  - **Highlights ductular reaction**
- **Amyloid A (SAA):**
  - **Stains as granular deposits in hepatocytes in inflammatory HCA**
  - ***Note: CRP (C-reactive protein) also an excellent marker for inflammatory HCA***

Well-Differentiated Hepatocellular Lesions: Benign or Malignant?

Differential Diagnosis:

FNH, HCA, HCC

Stains: Do these help?

GS, K7, SAA

Well-differentiated Hepatocellular Neoplasm Immunostains: Benign Vs. Malignant

- **Glutamine synthetase**
  - Present in centrizonal hepatocytes in normal liver, adenoma, and map-like staining in FNH
  - **Variable intensity and pattern in HCC**
- **K7**
  - Highlights ductular reaction in FNH, HCA
  - **K7 may stay acini, cholestatic areas, patchy staining**
- **Amyloid A (SAA)**
  - Granular deposits in hepatocytes in inflammatory HCA
  - **Variable staining in HCC**

Well-differentiated Hepatocellular neoplasms
Immunostains: Benign vs. Malignant

Differential diagnosis: HCA, FNH, HCC

What about these stains?

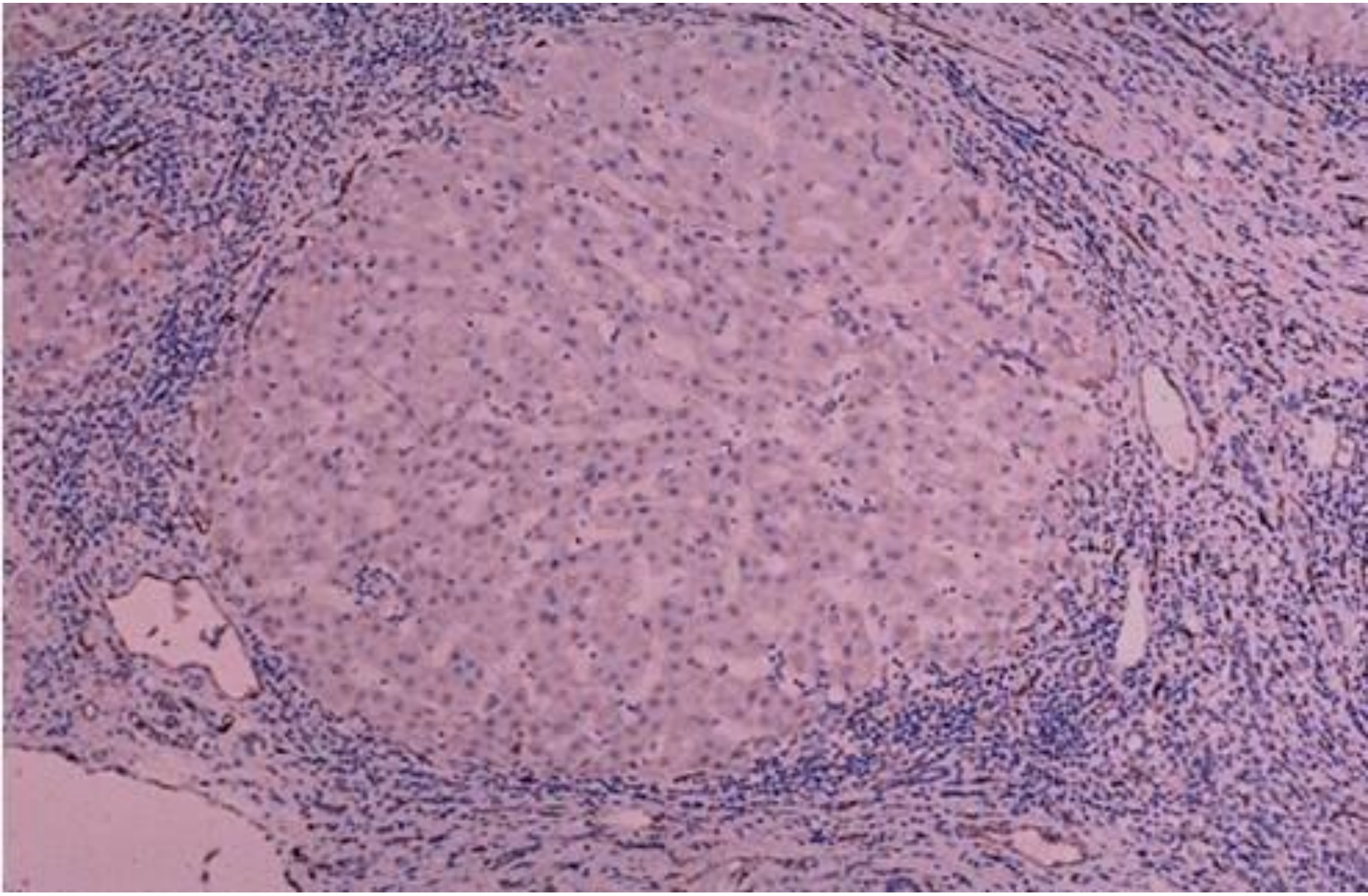
-CD34 ??

-Glypican-3 ??

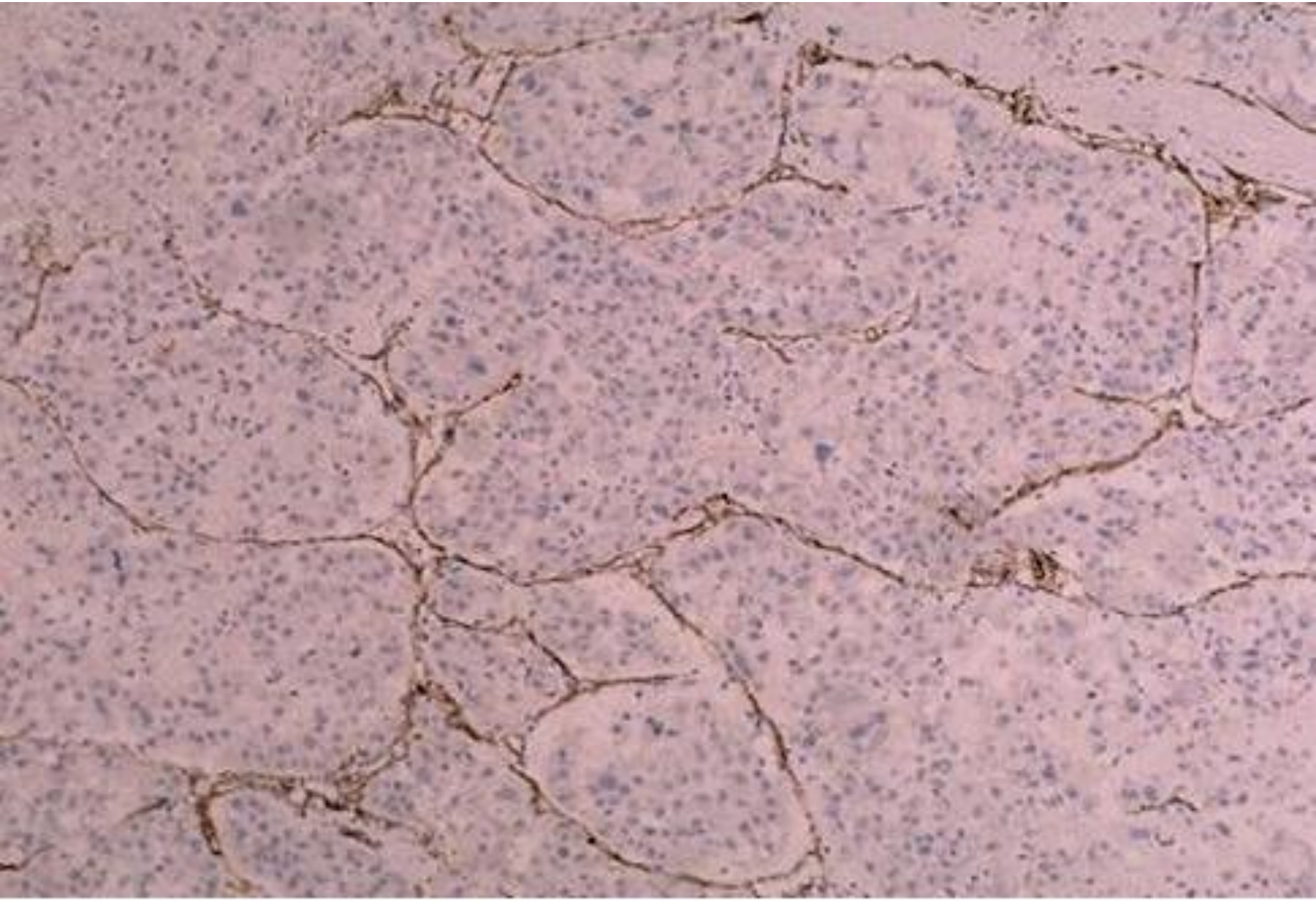
CD 34

- Does not stain normal sinusoidal endothelium
- *Stains endothelial lining of trabeculae of HCC and other lesions with increased arterial blood flow*
- Indicates “capillarization” of sinusoids

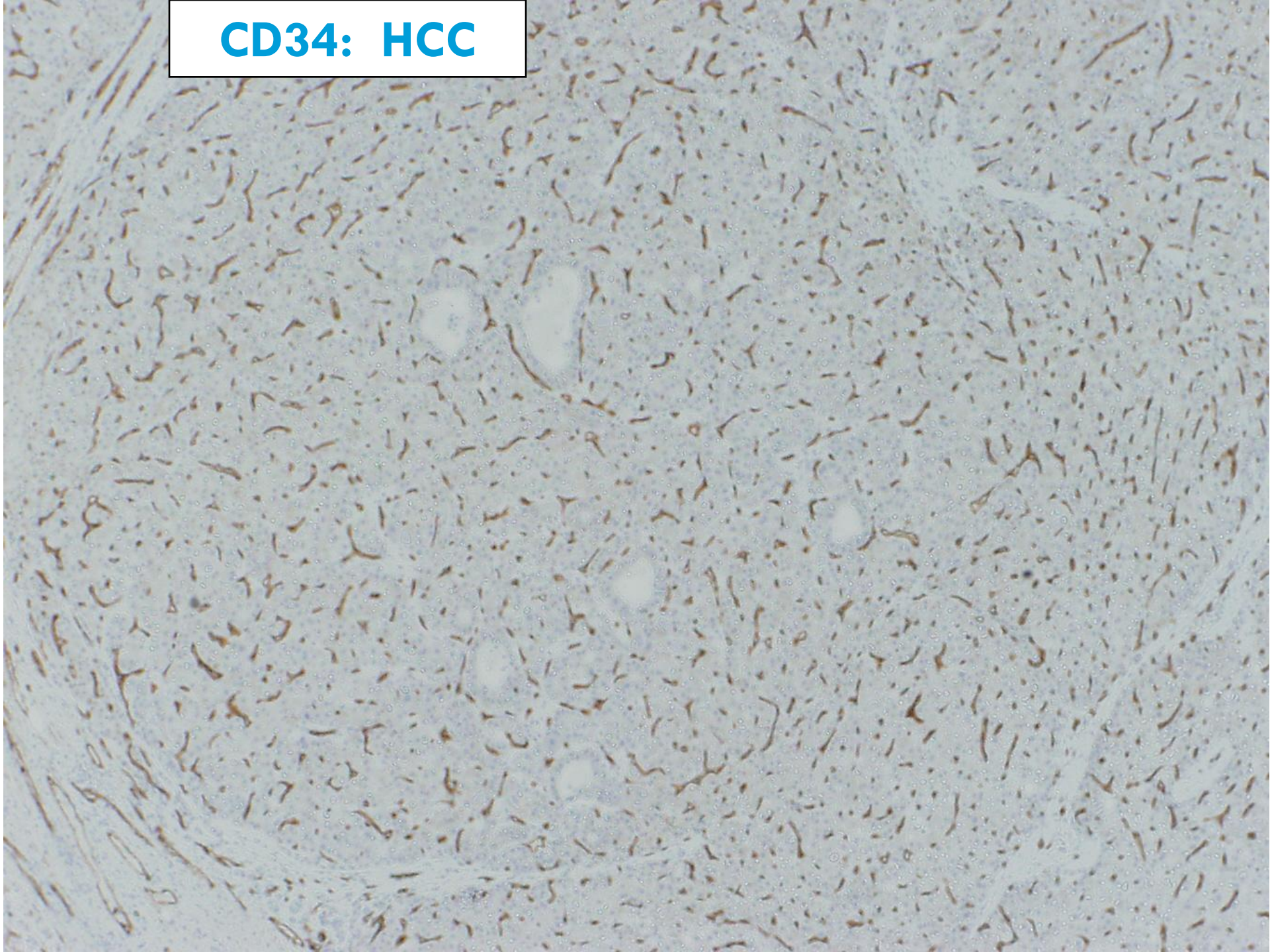
CD-34: Pattern most often seen in cirrhosis



CD-34 in HCC



CD34: HCC

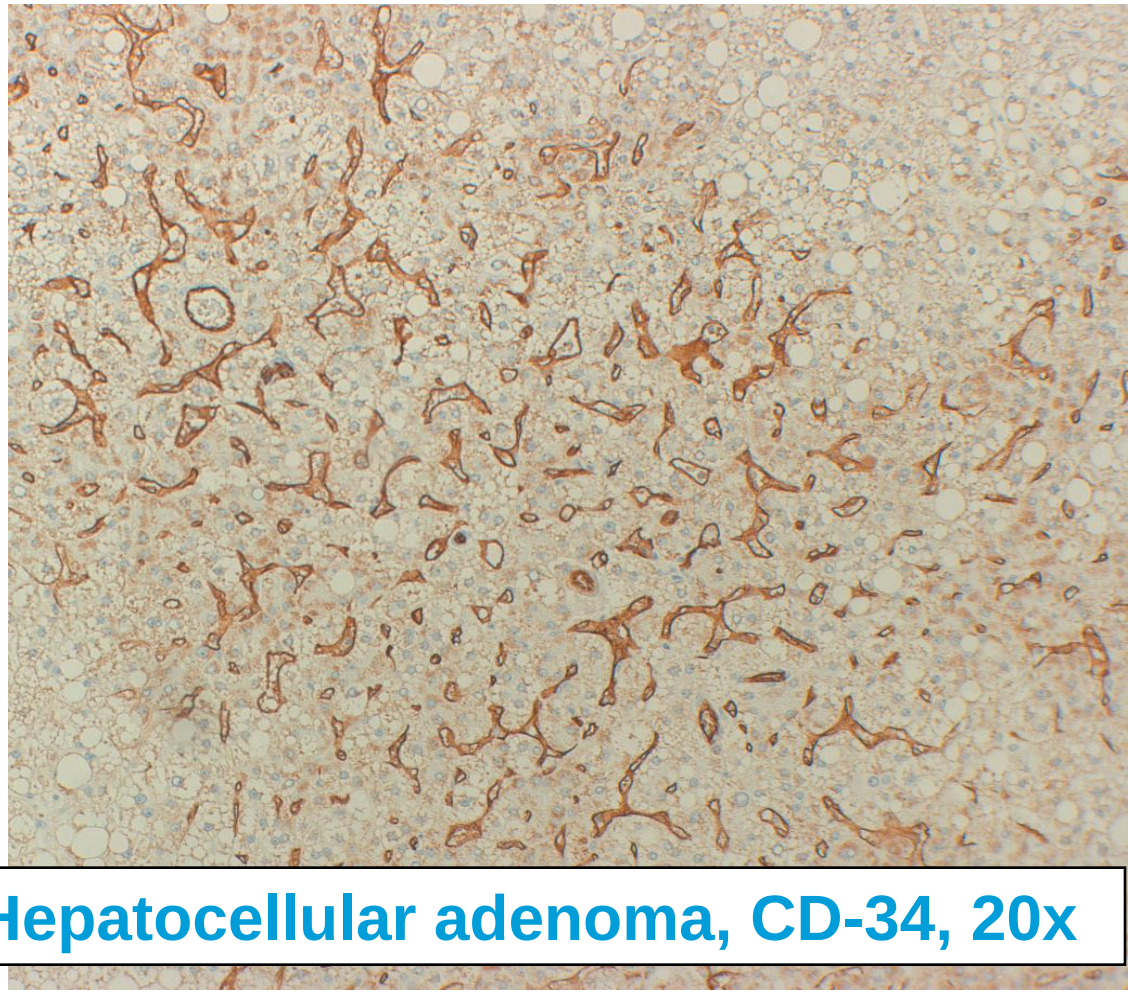


Immunostains: CD 34

Problem:

Also positive in:

Many adenomas, FNH, and high grade dysplastic nodules as well as some cases of cirrhosis



Hepatocellular adenoma, CD-34, 20x

Immunostains: CD34

- May help confirm presence of neoplasm
- Often + in HCC; can help in determining cell plate width
- Not commonly seen in nonhepatocellular tumors
- *NOT very useful to differentiate benign hepatocellular tumors from very well differentiated HCC*

Well-differentiated Hepatocellular Neoplasms: Immunostains

Benign vs. Malignant (HCA, FNH, HCC)

Glypican-3

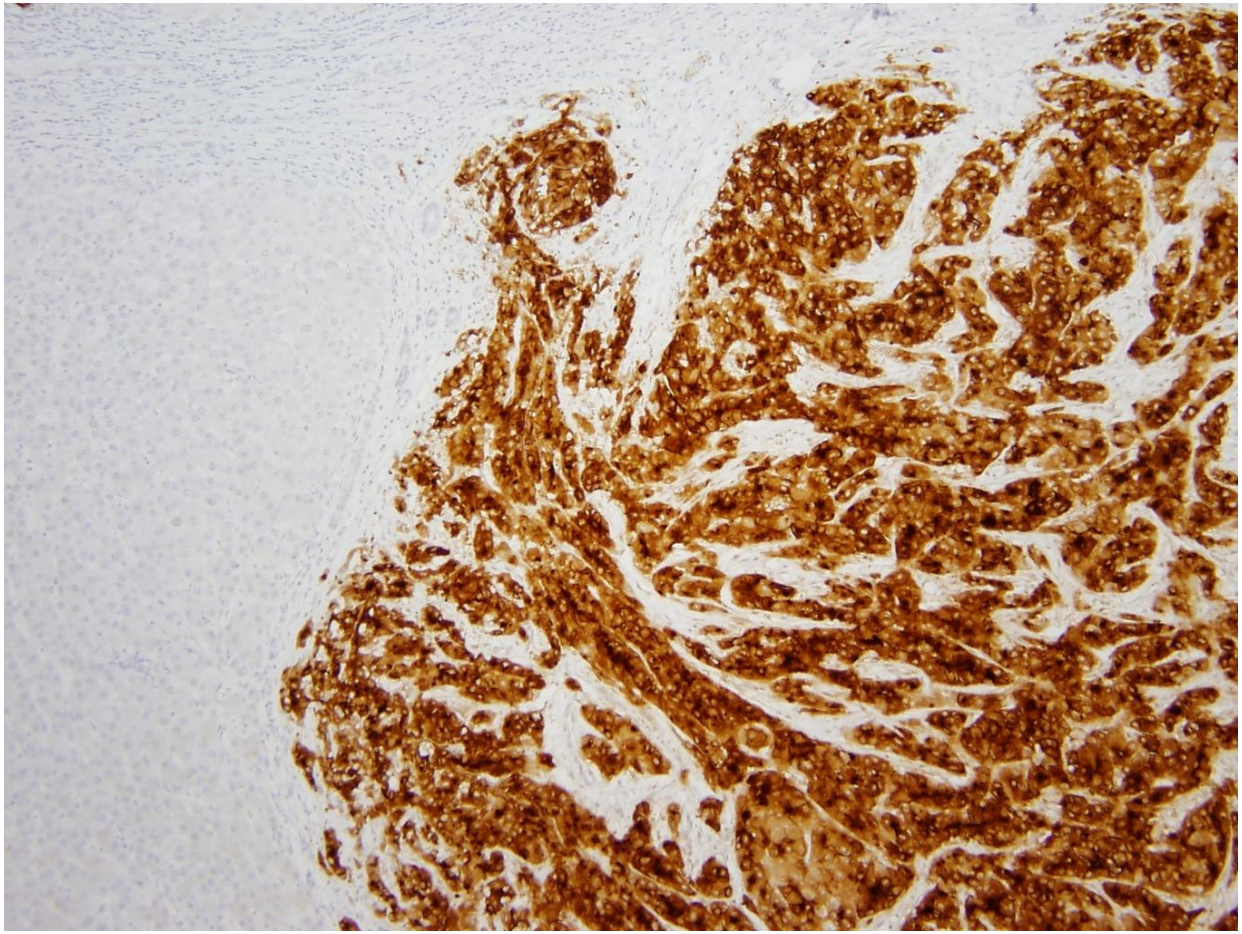
Negative in FNH and HCA

PROBLEM: no staining of most very well-differentiated HCCs, so not very helpful in distinguishing from FNH and HCA from WDHCC

Gly-3 in HCC

Cytoplasmic staining

Stromal invasion



Well-differentiated Hepatocellular Neoplasms: Immunostains

- **Benign vs. Malignant (HCA, FNH, WDHCC)**
  - CD34: not too helpful
  - Glypican-3: not too helpful for WDHCC
- **Reticulin stain abnormalities remain an accepted criteria but beware of overlapping features**
- **Chromosomal abnormalities may prove to be helpful (??)**
- **Other markers of the future (??): Heat-shock protein, metalloproteinases, other proteins?**

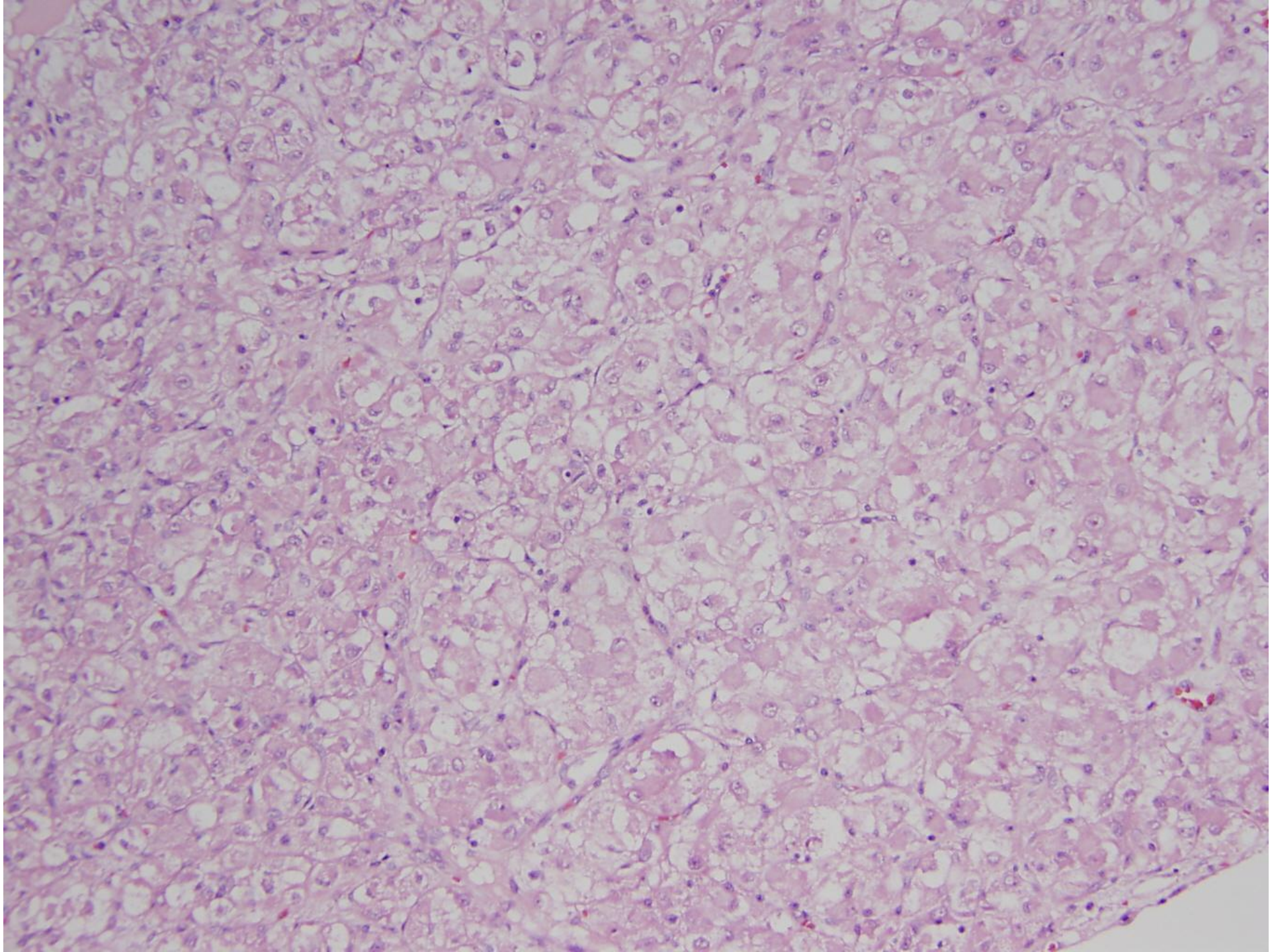
Other problems

Differential Diagnosis:

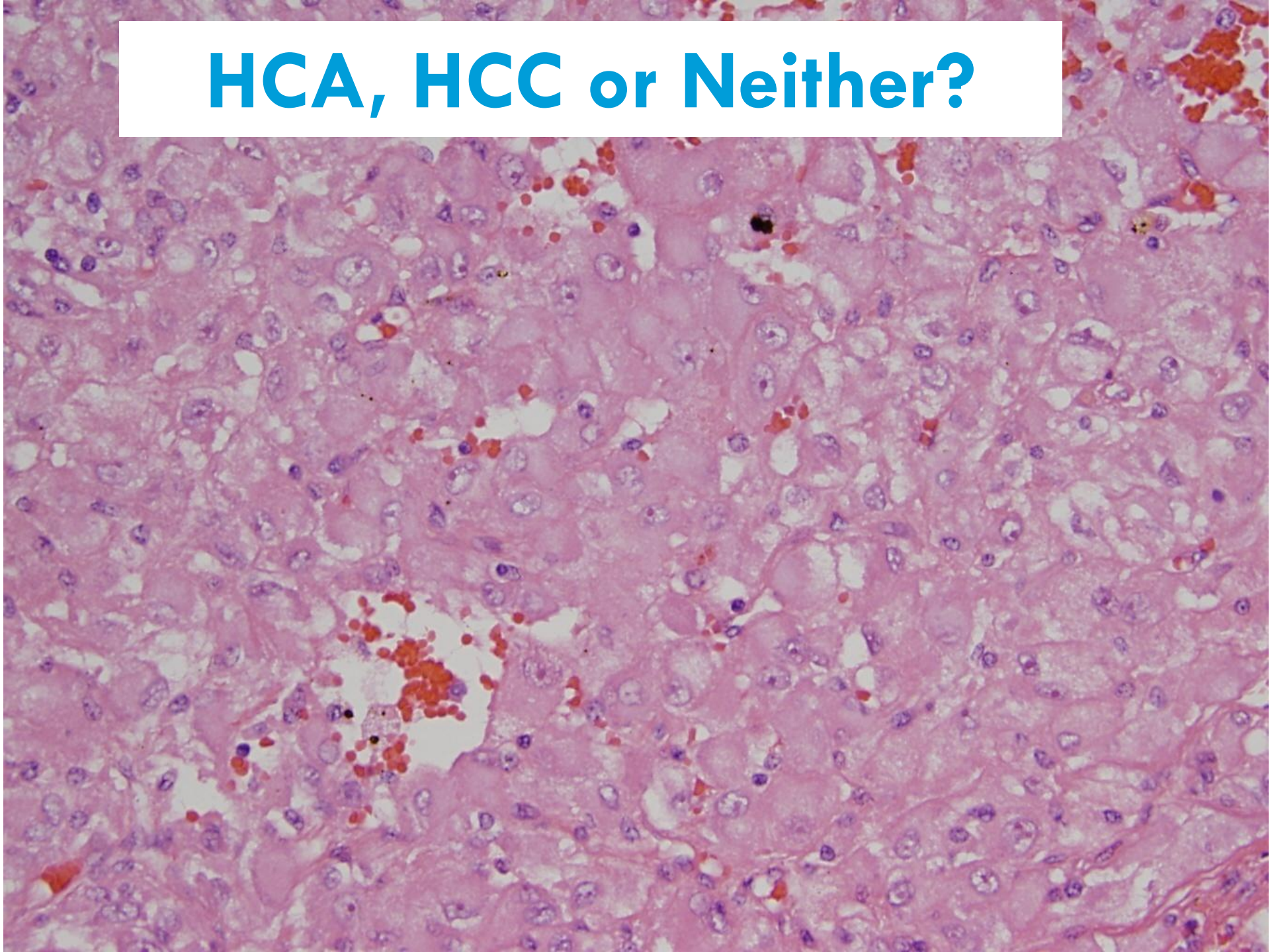
Well-Differentiated hepatocellular lesion or not?

Problem Case

- 37-year-old woman
- 11 cm pedunculated mass
- No cirrhosis or other risk factors for HCC
- Mass noted during routine gynecologic exam, no symptoms

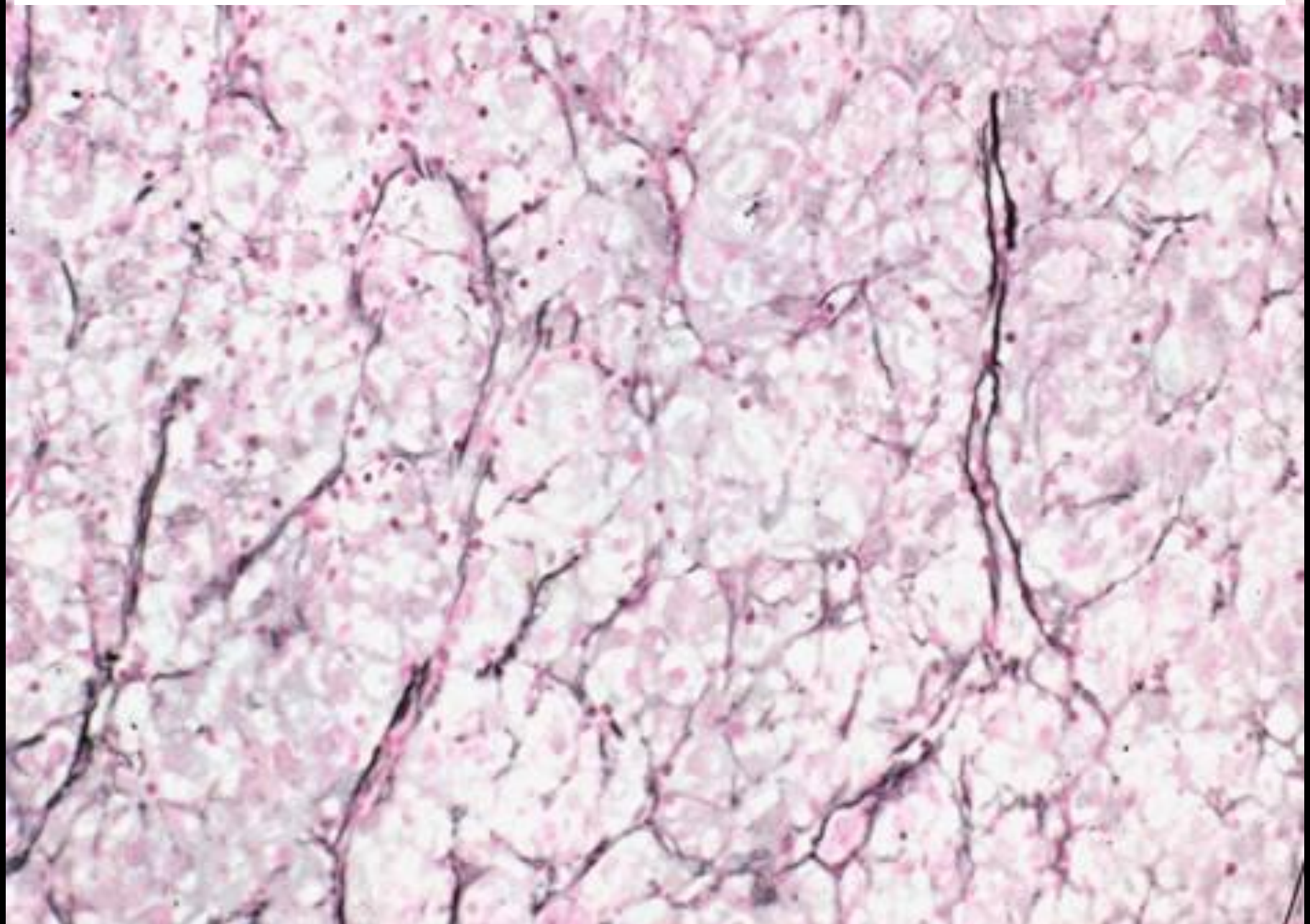


HCA, HCC or Neither?

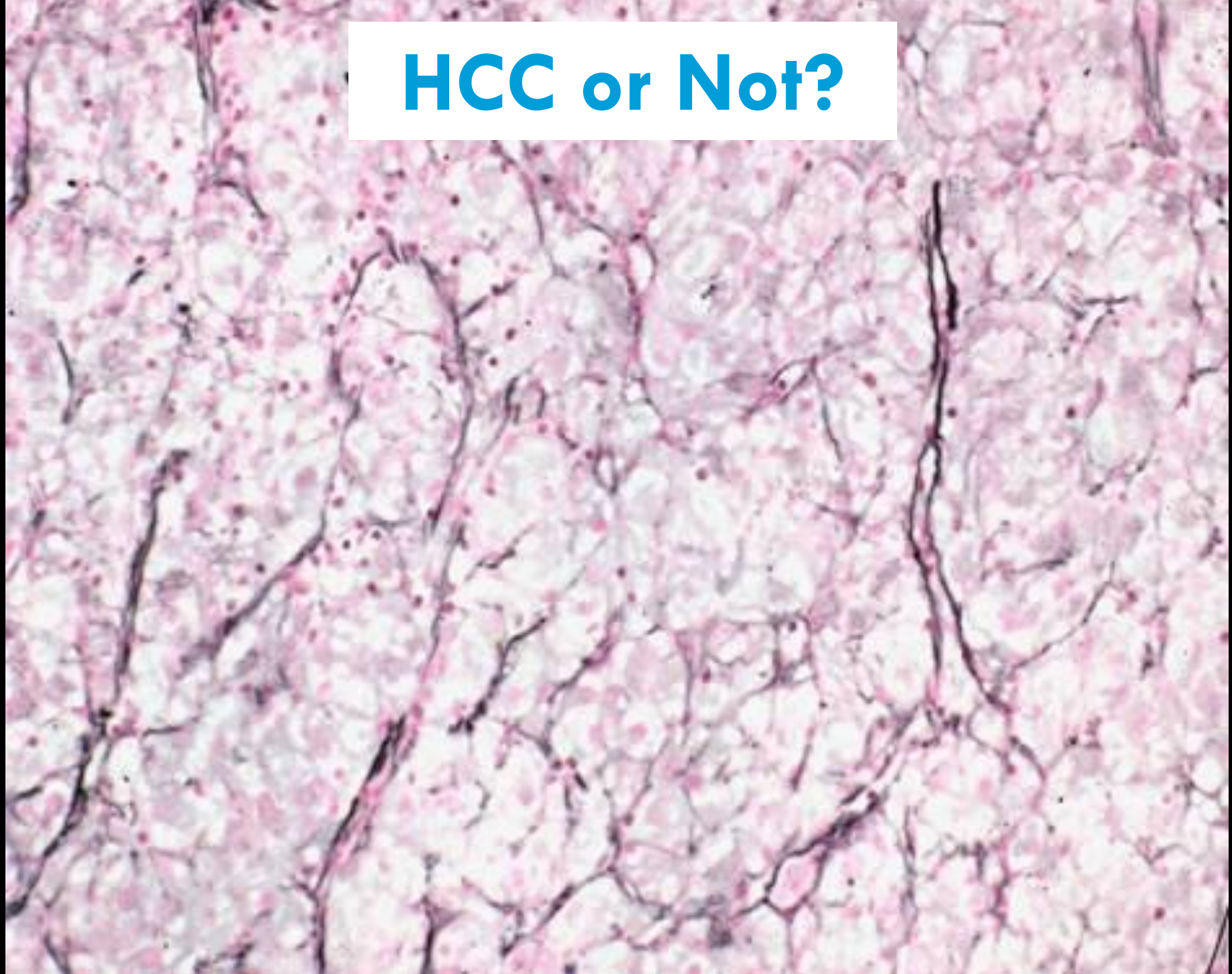


Reticulin Stain

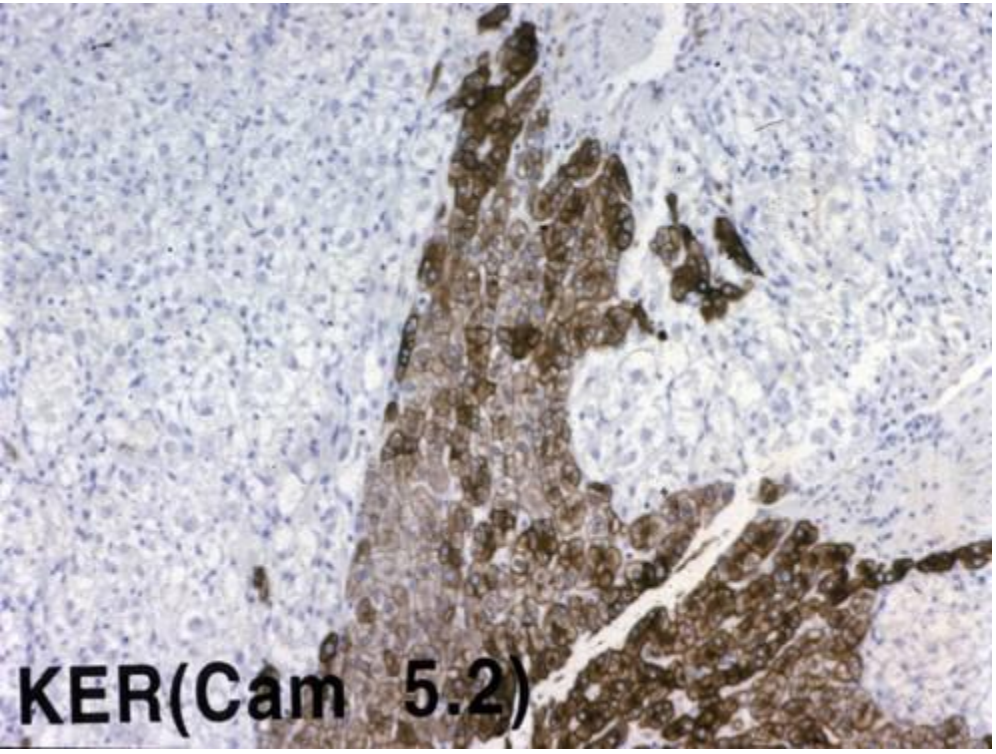
Reticulin Stain: too much loss for HCA

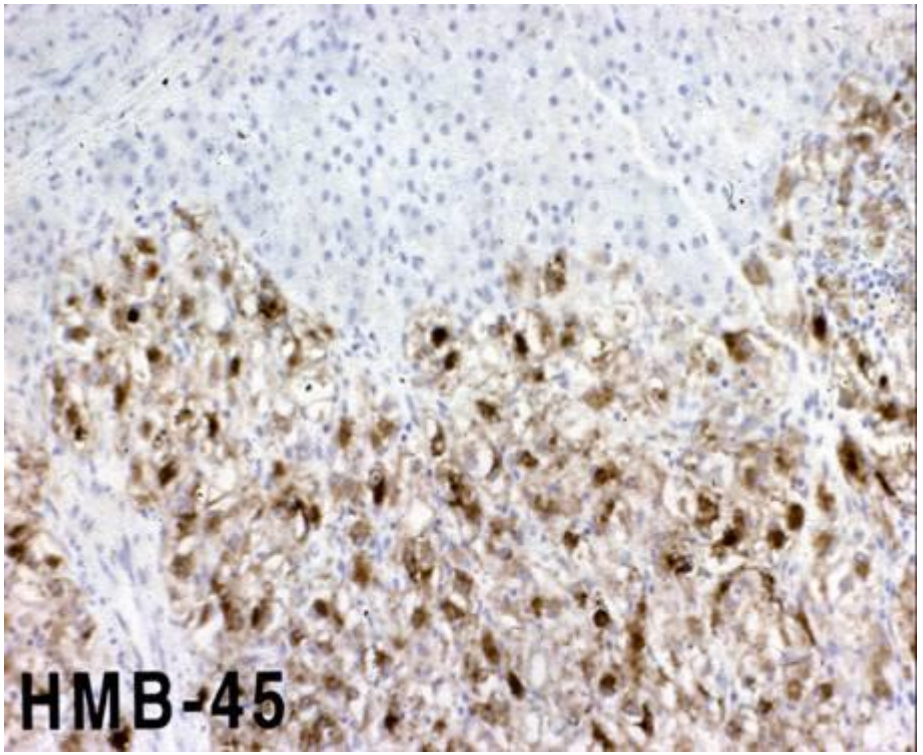


HCC or Not?

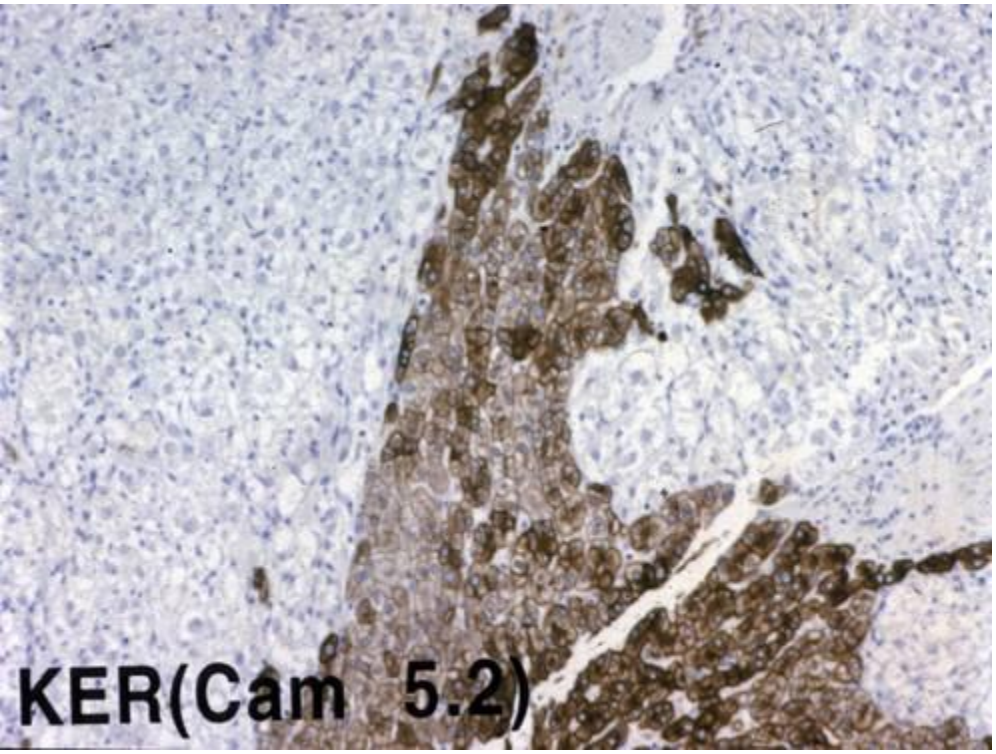


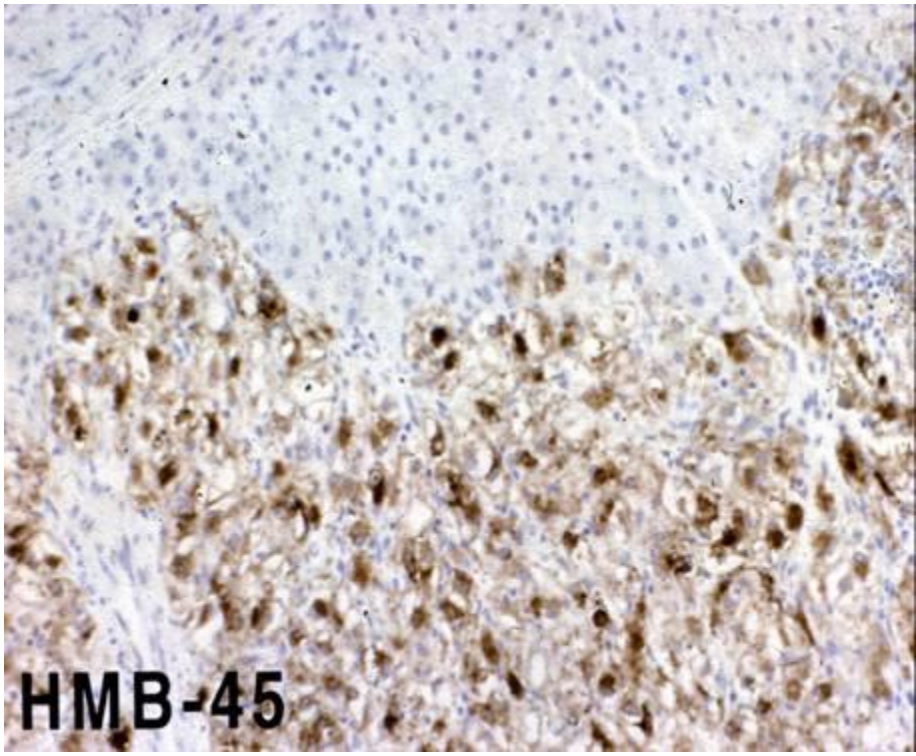
Keratin and HMB-45





Angiomyolipoma, epithelioid variant





Other problem tumors

Non-cirrhotic liver: Beware of epithelioid tumors that could mimic hepatocellular lesions

Immunohistochemistry may be helpful to differentiate from hepatocytic tumors

- **Angiomyolipoma → HMB-45, SMA**
- **Melanoma → S100, HMB-45**
- **Vascular tumors (angiosarcoma, epithelioid hemangioendothelioma) → CD31, CD34, Fli1**
- **Gastrointestinal Stromal Tumor (GIST) → CD117 (C-kit), DOG1, CD34**

Summary

- Cirrhotic liver
  - MRN, HGDN versus Well-differentiated HCC
    - More pronounced small or large cell change
    - Reticulin framework loss or abnormal architecture
    - Invasion, stromal or lobular

    Notable variant: Cirrhosis-like HCC
- Non-cirrhotic liver
  - FNH, HCA, HCC
    - Similar histologic issues for HCC as above
    - Beware of some architectural overlapping features
    - Glutamine Synthetase, Serum Amyloid A for FNH/HCA
  - Lesions mimicking HCC: Immunohistochemistry