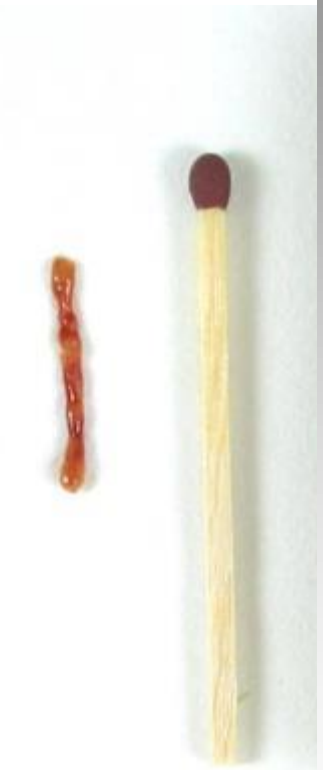
The what, why and how of reporting medical liver biopsies

Judy Wyatt

June 2020

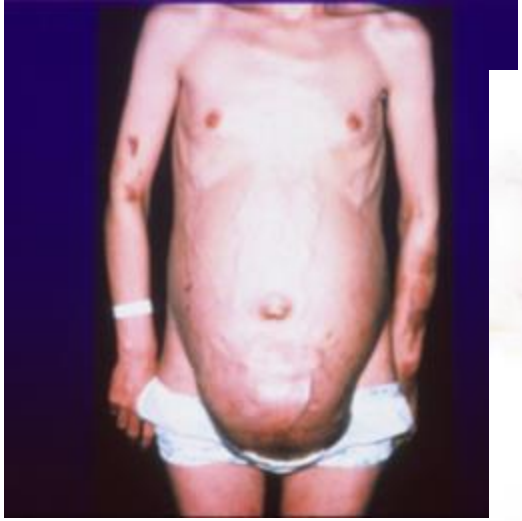
What?

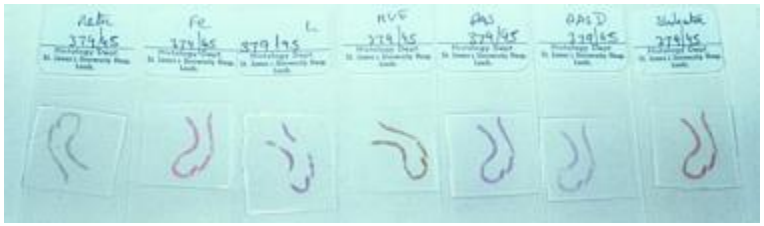
Percutaneous liver biopsy

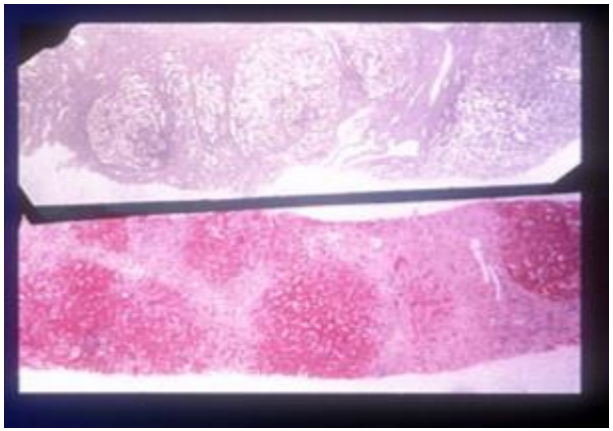


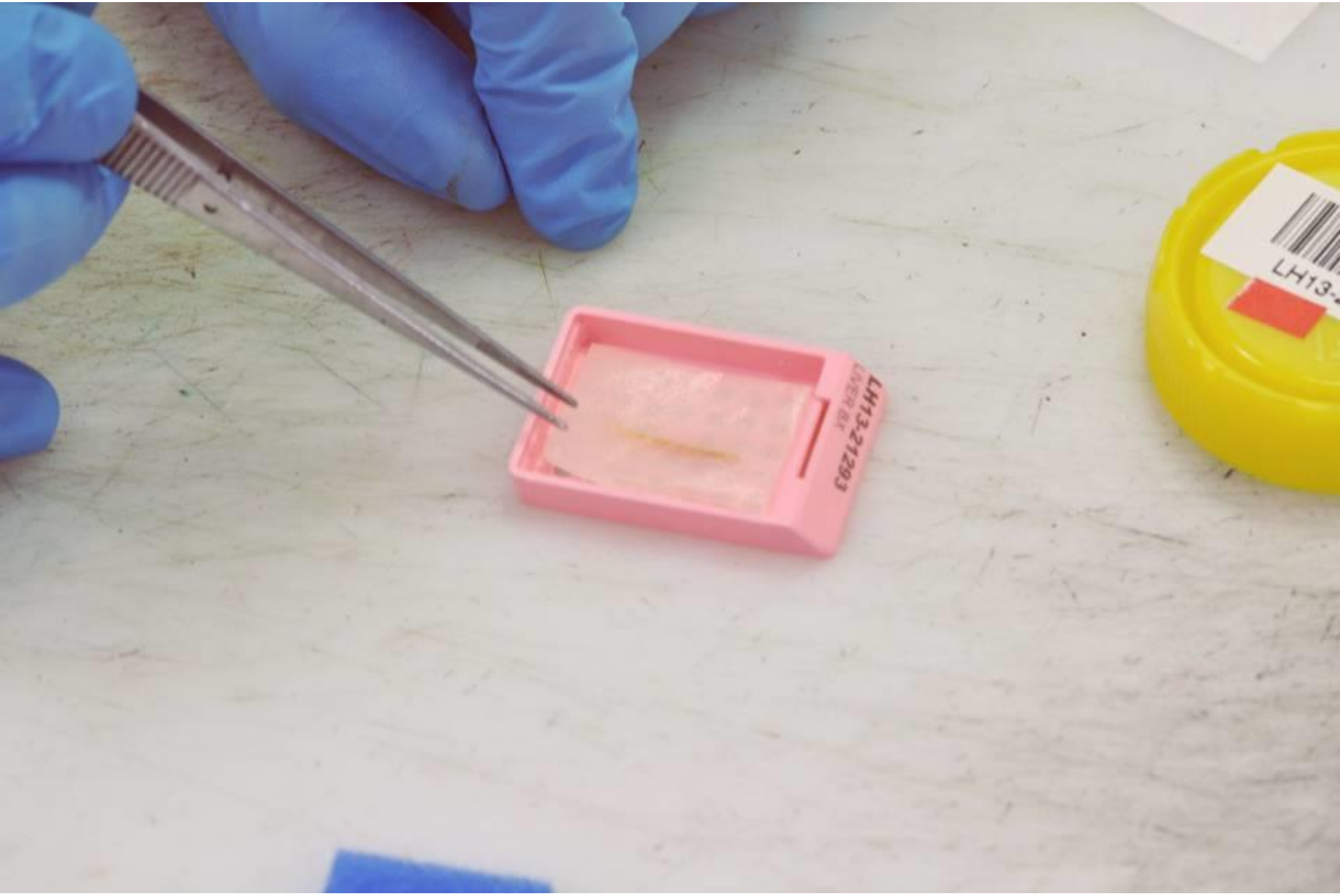
1/50,000th of the liver

- Essential not to loose information during handling / processing of these small specimens





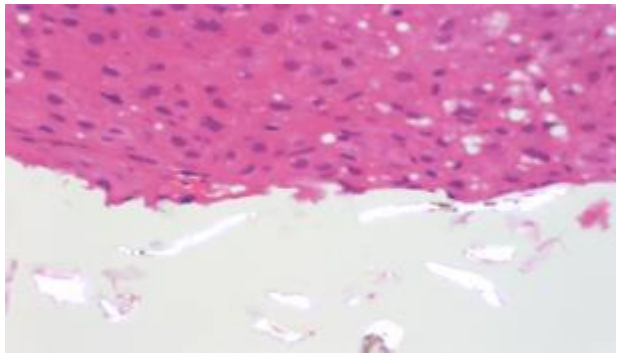



LH13-2129

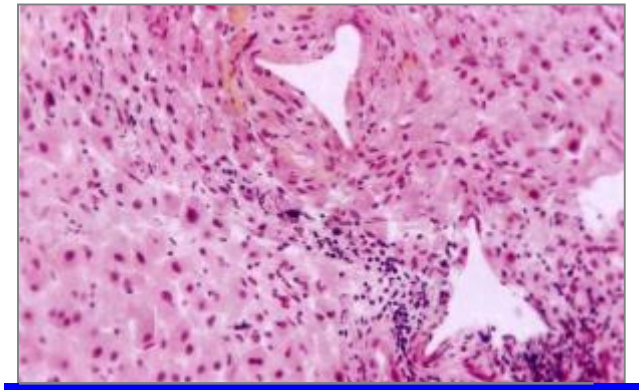
Handling the biopsy specimen

Liver biopsy specimens are best left floating in the fixative solution

⇨ **No blotting paper**



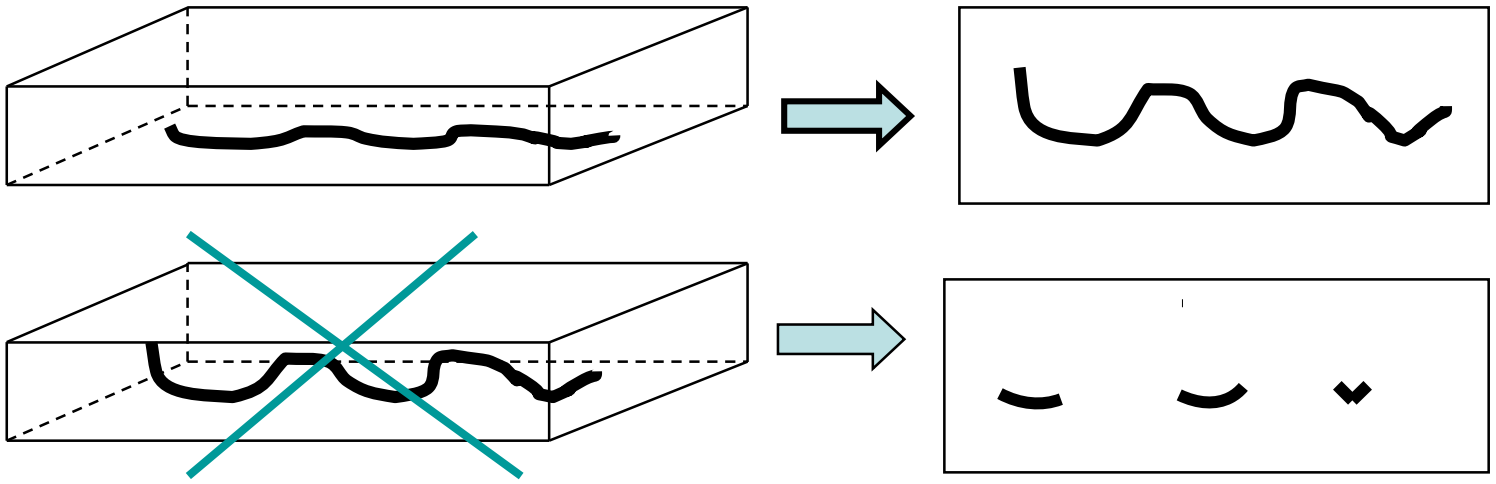
⇨ **Avoid rough packing between foam sponge**



⇨ **Use fine mesh cassettes or lens-paper wrapping**

Handling the biopsy specimen

Essential to embed specimen flat, especially when distorted, in order to get most of the core(s) cut by each microtome stroke



Paraffin wax blocks

Glass slides

The Royal College of Pathologists

Pathology: the science behind the cure

Tissue pathways for liver biopsies for the investigation of medical disease and for focal lesions

March 2014

Authors

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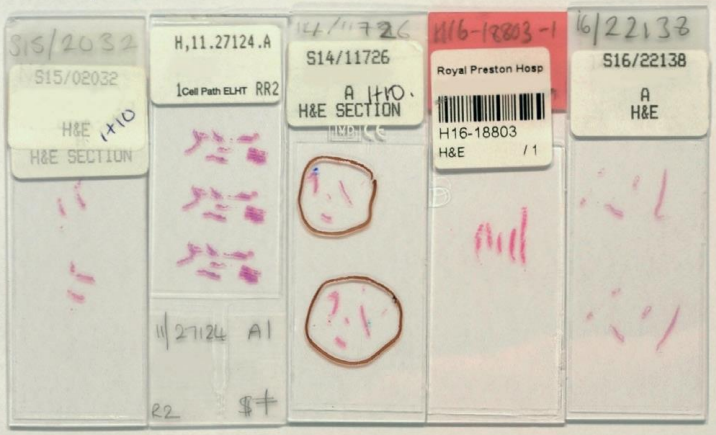
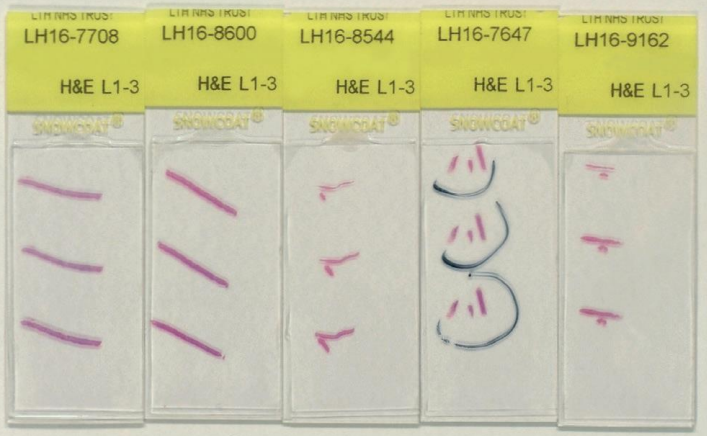
Unique document number	G064
Document name	Tissue pathways for liver biopsies for the investigation of medical disease and for focal lesions
Version number	2

A: biopsies from elsewhere for review



B: biopsies from previous needle, inconsistently adequate



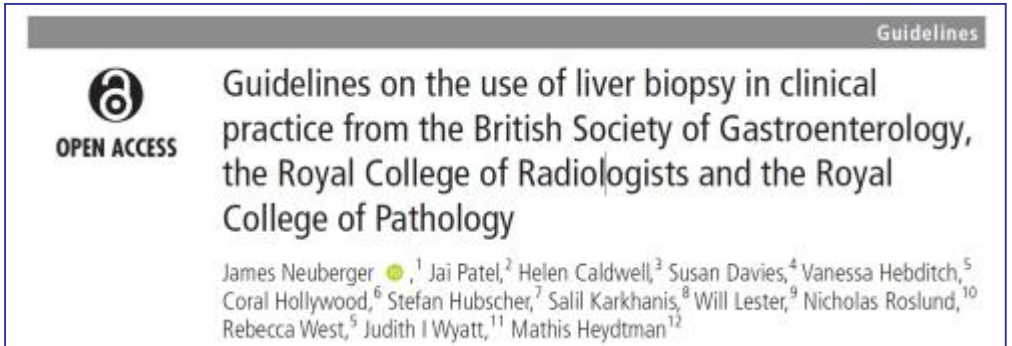
C: consistently good specimens from Biopince™

Recommendations for liver biopsy specimens

- Biopsies with <10 portal tracts underestimate disease severity (stage and grade)
- Biopsies <20mm underestimate fibrosis stage
- Biopsies with 18G needle are narrow and more likely to fragment and contain fewer portal tracts
- Avoid second pass if possible – although no clear evidence of risk

Recommend: 16G needle, core >20mm

Consider second pass if <20mm and definitely if <10mm.


Guidelines

OPEN ACCESS

Guidelines on the use of liver biopsy in clinical practice from the British Society of Gastroenterology, the Royal College of Radiologists and the Royal College of Pathology

James Neuberger ,[1] Jai Patel,[2] Helen Caldwell,[3] Susan Davies,[4] Vanessa Hebditch,[5] Coral Hollywood,[6] Stefan Hubscher,[7] Salil Karkhanis,[8] Will Lester,[9] Nicholas Roslund,[10] Rebecca West,[5] Judith I Wyatt,[11] Mathis Heydtman[12]

Principles - How to handle the liver biopsy

⇨ Invasive technique - small specimen

⇨ High standard processing to avoid loss of information

⇨ Sensible use of available techniques

especially immunohistochemistry

⇨ Seek second opinion for challenging cases

(but limited assistance if poorly processed
+ paraffin block empty)

Importance of

Clinical awareness on the part of the pathologist

Clinical information given by clinicians

Interactive discussion between pathologist and clinician

Why?

These are biopsies taken for the investigation of diffuse parenchymal liver disease.

The common indications for biopsy include:

- persistent unexplained abnormality of liver biochemistry (abnormal 'liver function tests'), where there is
- no clear medical diagnosis after routine 'liver screen' investigations, or when
- such investigations result in more than one possible diagnosis

- assessment of severity/stage of a known disease, and to monitor change over time or with treatment.

- In liver centres, also acute liver failure and transplant biopsies.

Benefit > risk

Why? Reason for the biopsy -

Abnormality of liver biochemistry without known clinical diagnosis

- What could it be? Pathologist guides further investigation

△ Beware unqualified diagnosis of ‘chronic hepatitis’

Clinical details suggest one or more specific diagnosis

- Does histology support/exclude a diagnosis?
- May be more than one – which is dominant?

△ Beware – do not simply report a biopsy as ‘consistent’ with the clinical diagnosis without considering alternatives.

Risks of biopsy

- Death
  - 0.11% (Mayo clinic 1990,
  - 0.13-0.33% (UK DGH audit 1994)
  - 4/3500 (0.11%) Royal College of Radiologists UK Audit 2008
    (all deaths were in patients with biopsies of tumours)

  <1 in 10,000 for medical liver biopsies in the UK
  - based on linking HES data with death registry.

- Morbidity
  - Pain 30% (severe 3%)
  - Hypotension 3%
  - Significant haemorrhage <0.5%
  - Haematoma 23%
  - Haemobilia 0.05%
  - Puncture other viscera 0.01% - 0.1%

Guidelines on the use of liver biopsy in clinical practice, BSG, RCR, RCPath April 2020

Pattern recognition in medical liver diseases

Acute liver disease – high transaminases and/or bilirubin

– no fibrosis, normal vascular relations

similar distance between portal tracts and hepatic veins

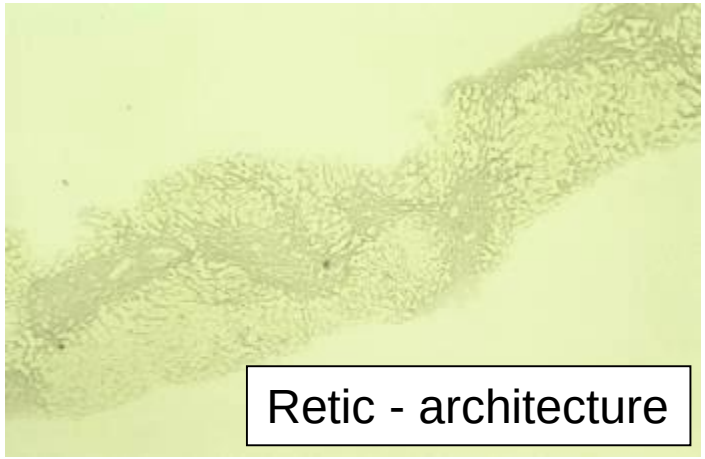
- hepatitis - inflammatory
- cholestasis - bilirubinostasis
- vascular/toxic – ischaemic

Four patterns of chronic liver disease

- Chronic hepatitis
- Chronic biliary disease
- Fatty liver disease
- Vascular disease

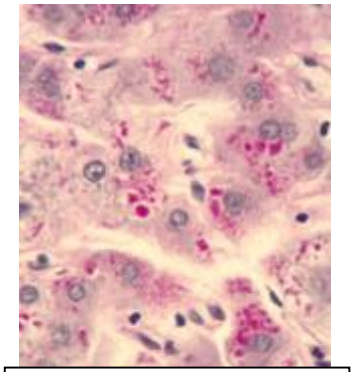
Chronic hepatitis

- Portal expansion and fibrosis
- Portal tract chronic inflammatory infiltrate
- Interface hepatitis
- Absence of features of biliary disease

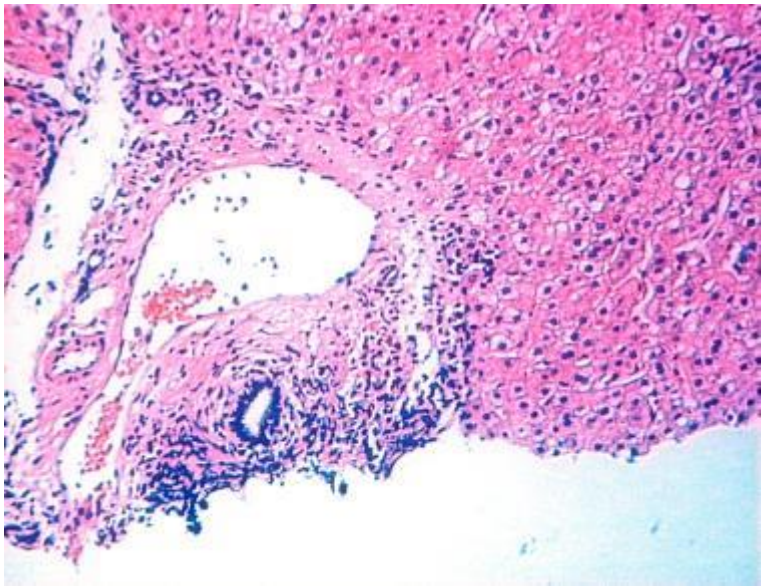


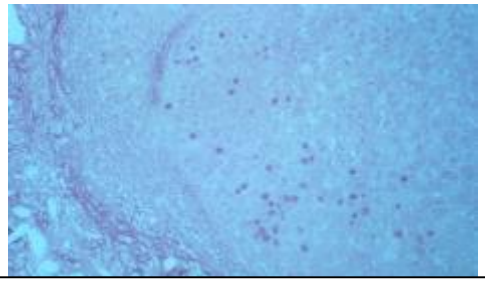
Retic - architecture

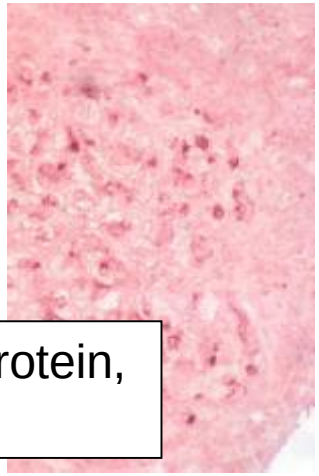
Perl's - iron



PASD – a1atd





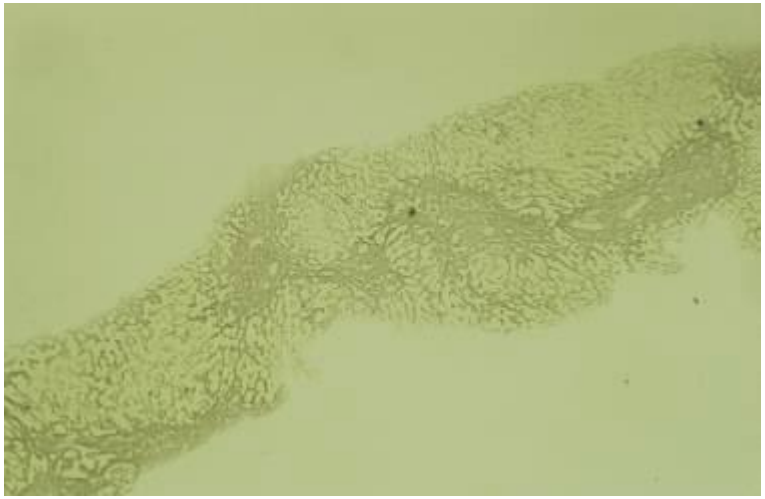


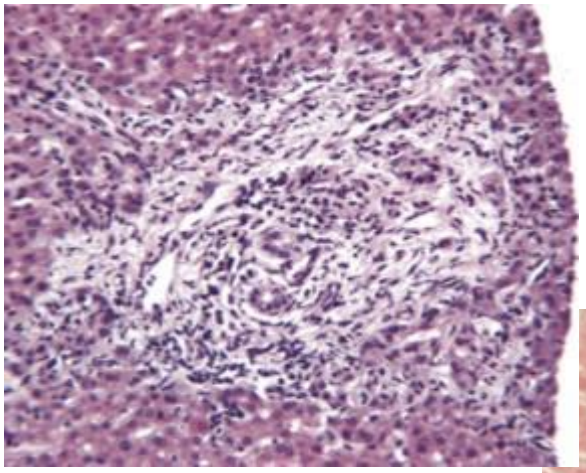
Shikata – copper-binding protein, Hepatitis B sAg, elastic

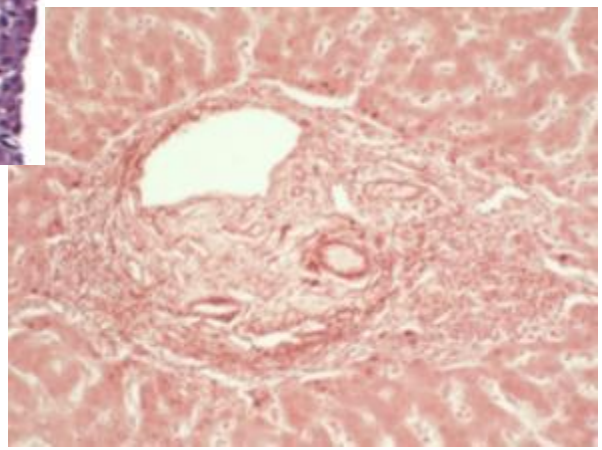
Chronic cholestasis

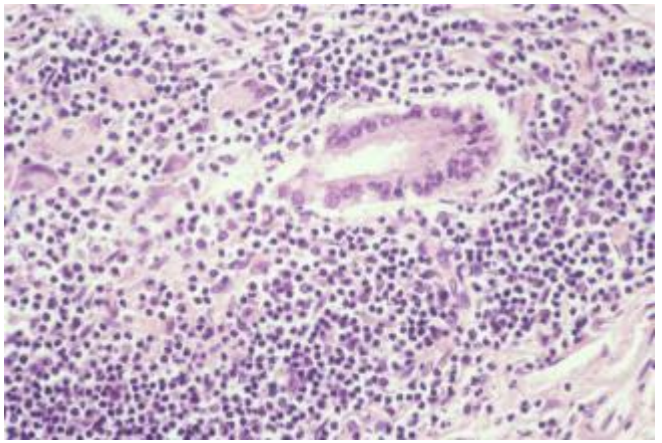
- Portal fibrosis
- Ductular proliferation
- Copper associated protein

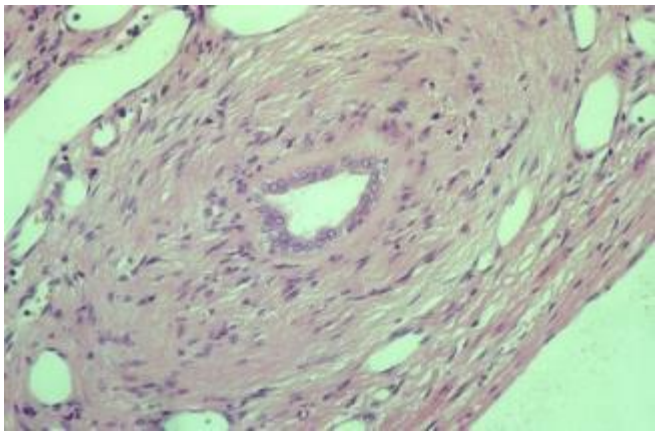
- Bile duct lesions
- reduction in bile duct number





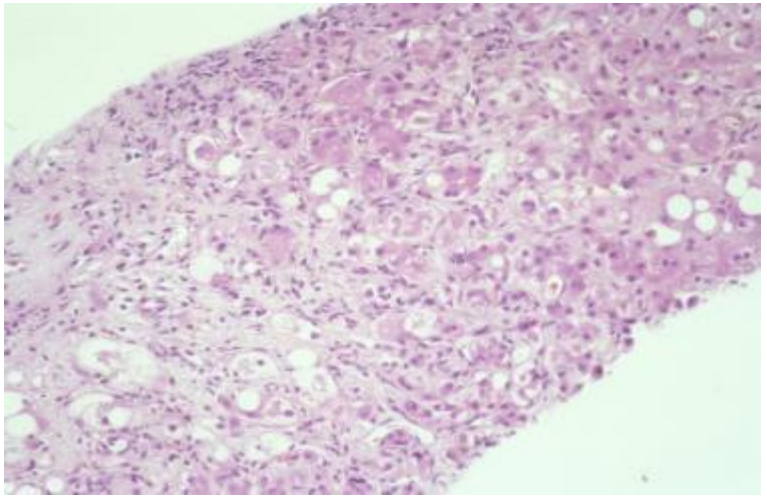


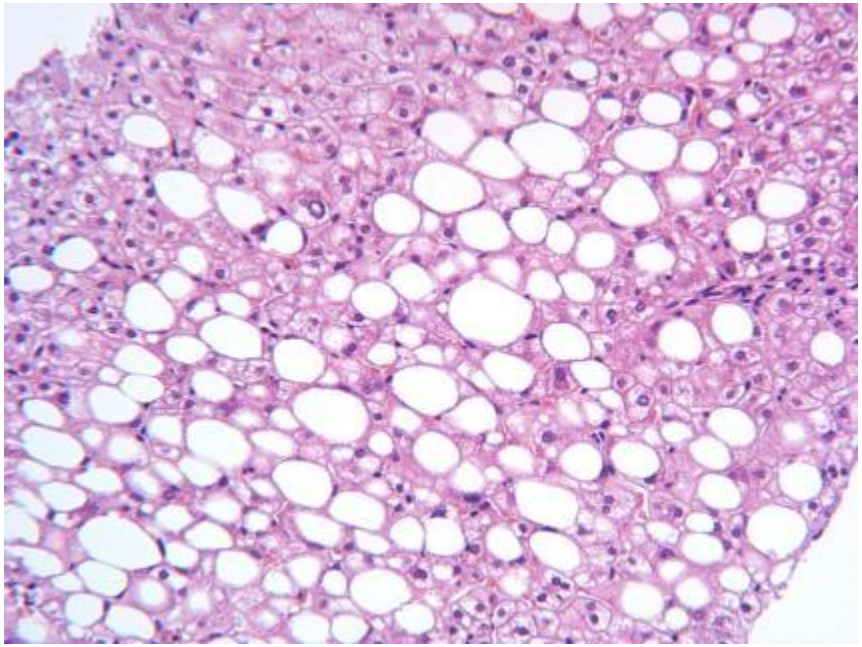


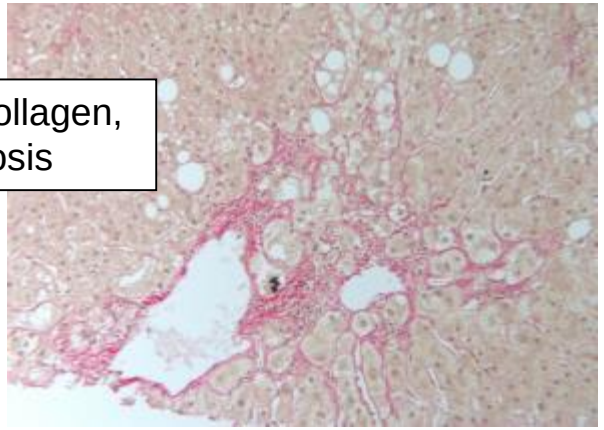


Fatty liver disease

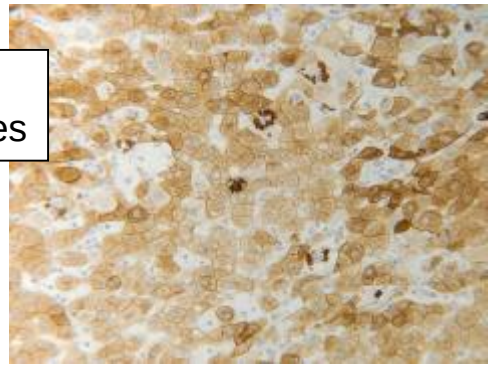
- micro/macro vesicular fat
- Variable but mild portal inflammation
- ? Other features of steatohepatitis
  - ballooning, Mallory bodies,
  - lobular inflammation
  - pericellular fibrosis







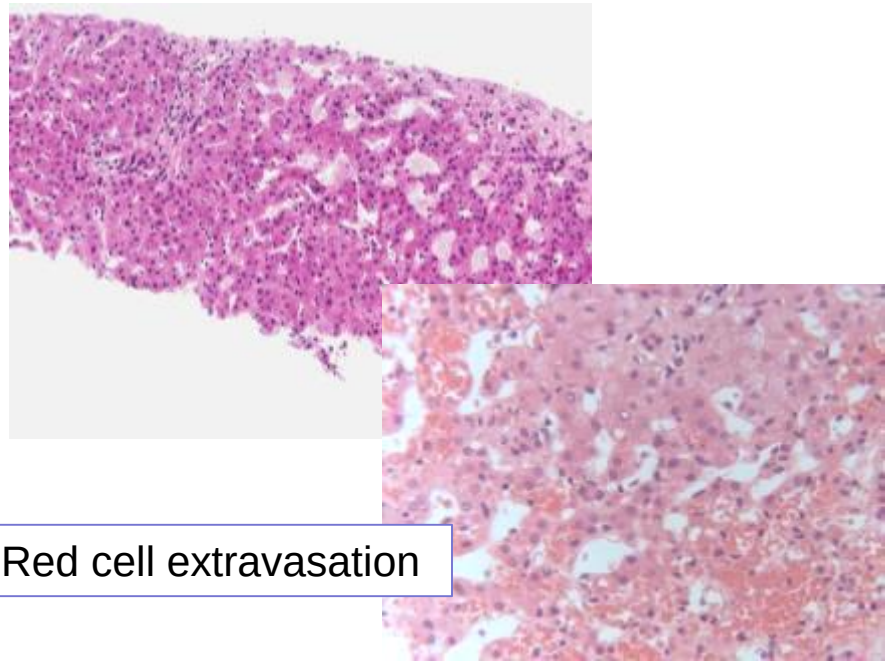
Van Gieson – collagen,
Pericellular fibrosis



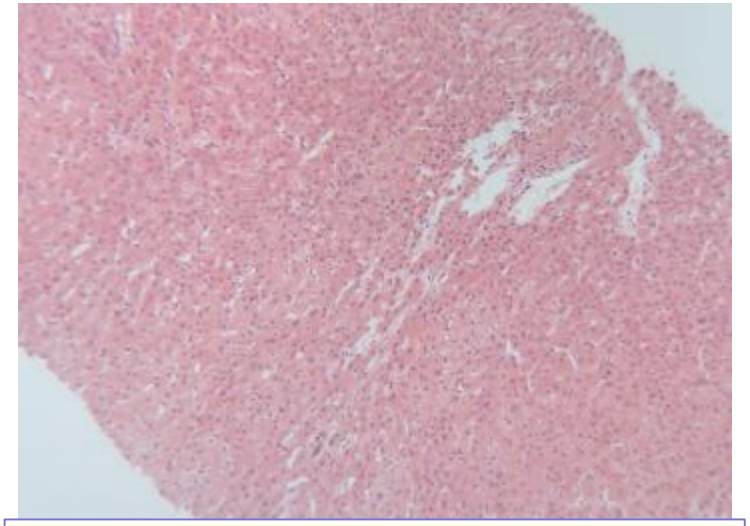
CK8/18,
Mallory bodies

Vascular disease

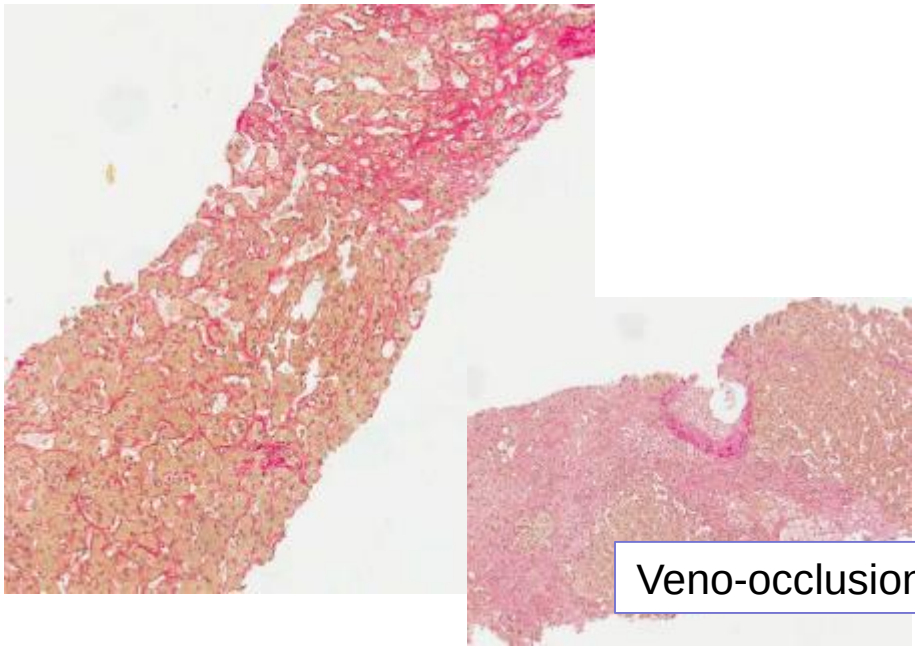
- No inflammation
- Congestion of sinusoids
  +/- red cell extravasation
- Sinusoidal fibrosis associated with small size hepatocytes
- Nodular regeneration without fibrosis
- zone 3 based fibrosis



Red cell extravasation



Nodular regeneration without fibrosis



Veno-occlusion

Liver special stains

Usual panel

Architecture:

- Retic
- van Gieson
- Shikata

Hepatocytes:

- PAS, PASD,
- Shikata

Pigment:

- Perl’s

Usual panel

Architecture:

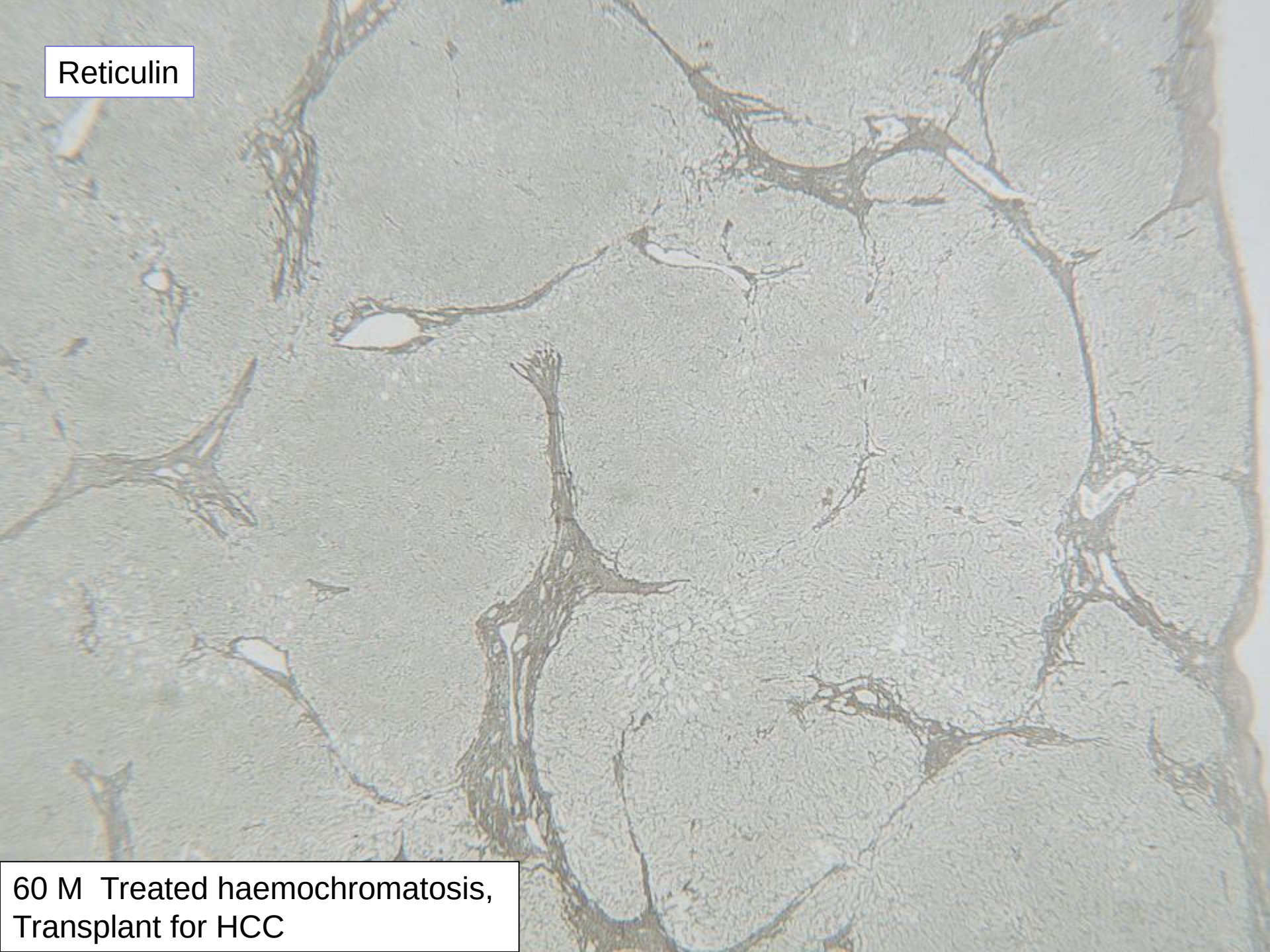
- Retic – liver cell plates, stage of chronic liver disease
- van Gieson – mature collagen, acute v. chronic liver disease, hepatic veins
- Shikata – elastic, in vessels and long standing fibrosis

Hepatocytes:

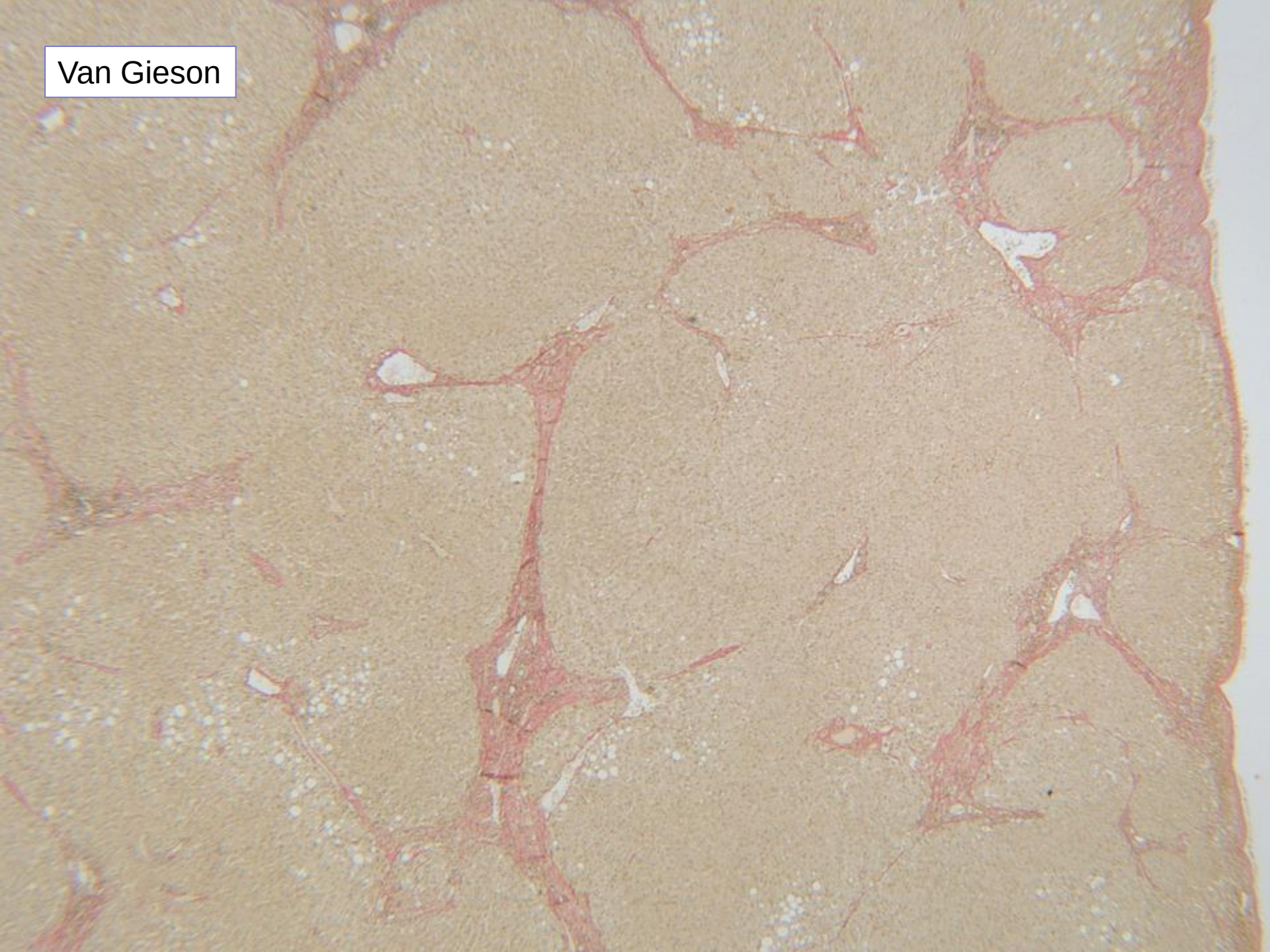
- PAS, PASD,
- Shikata

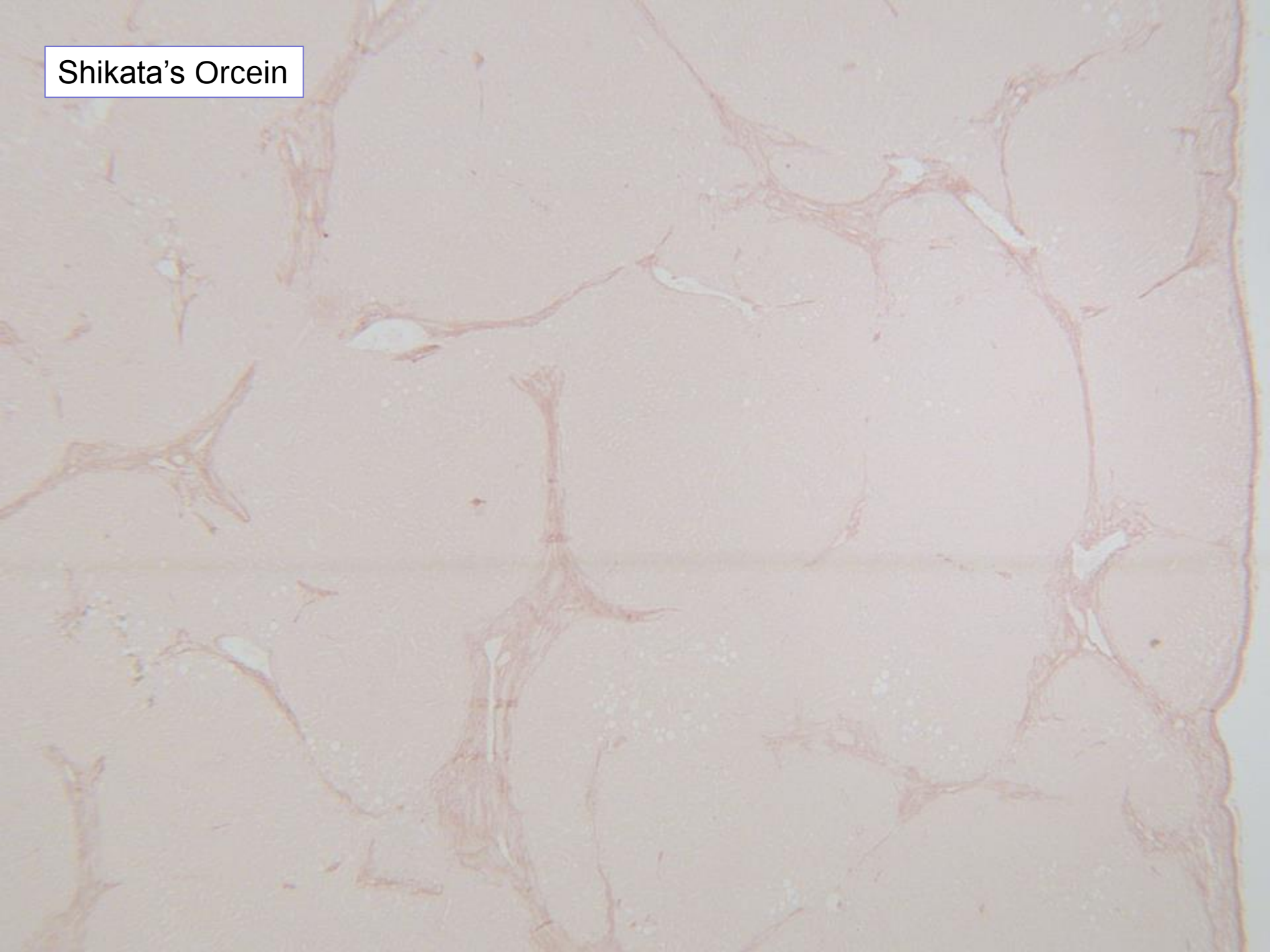
Pigment:

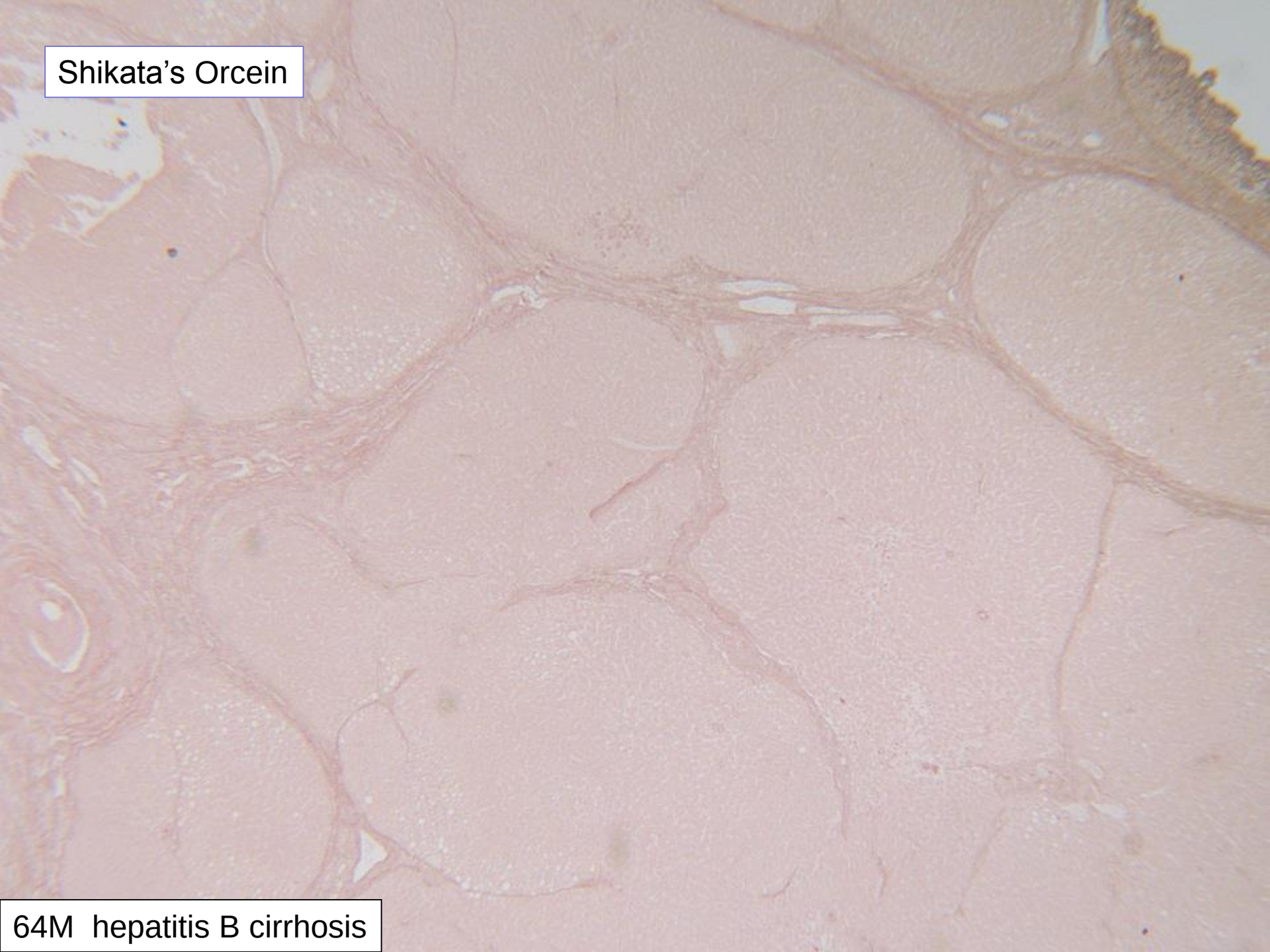
- Perl’s

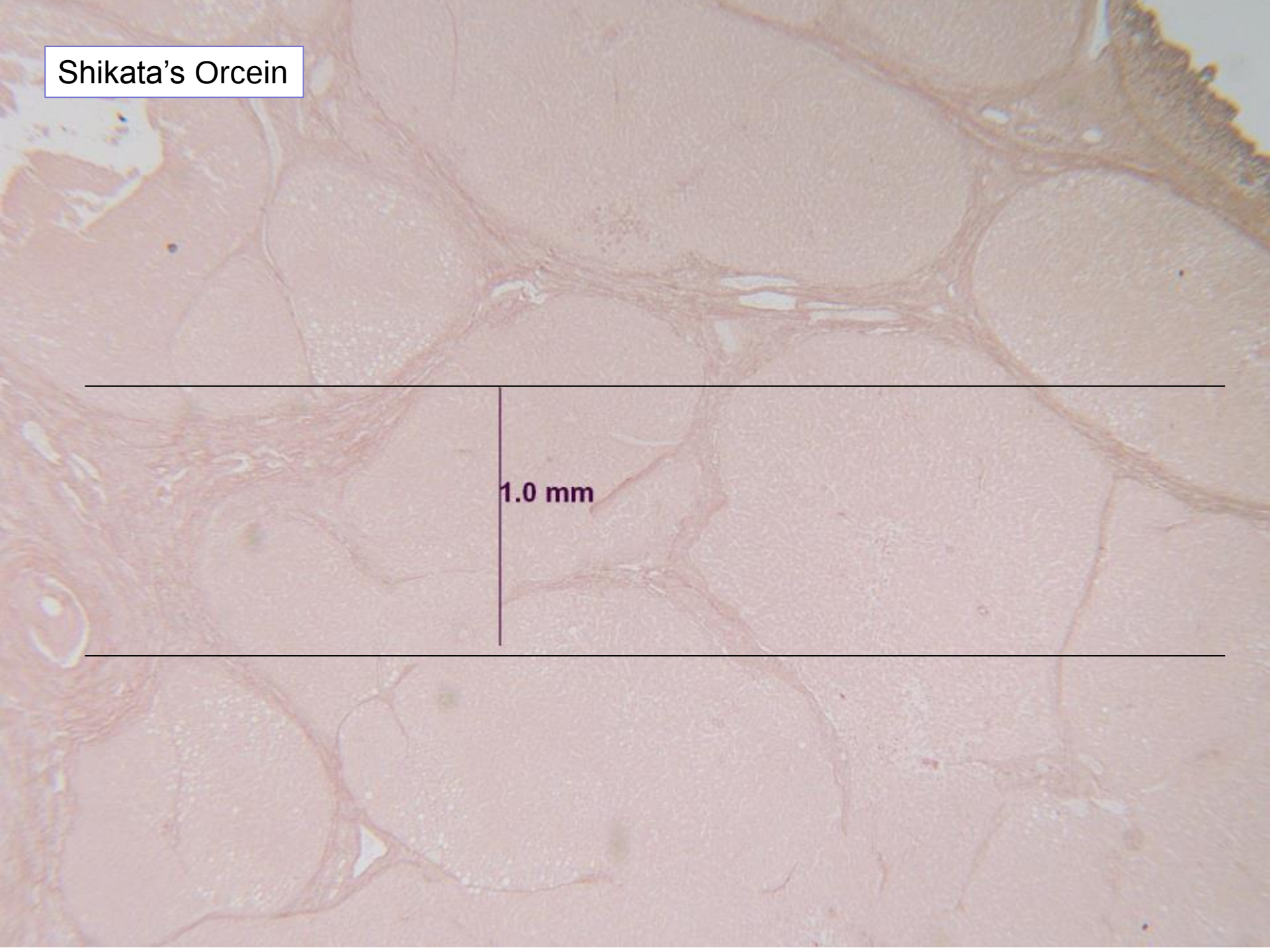


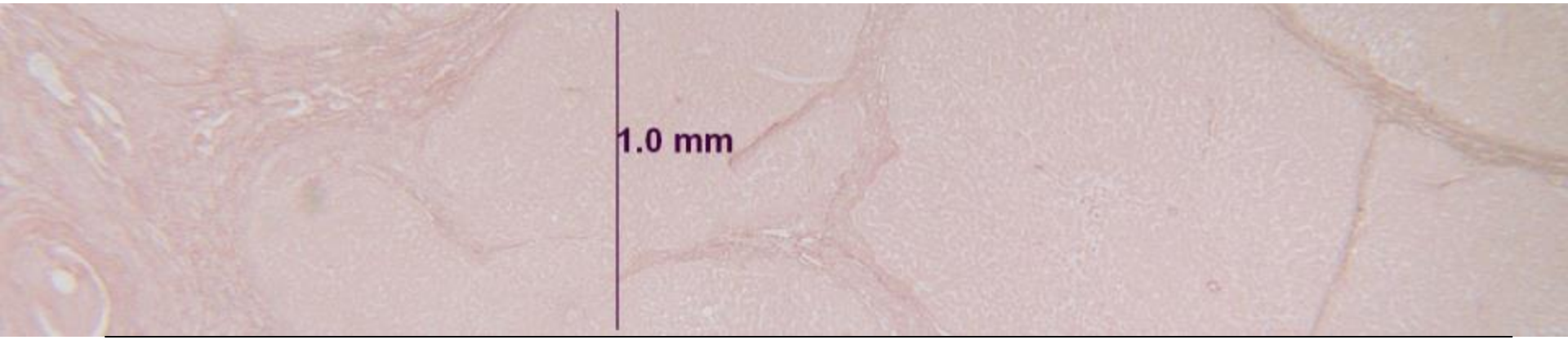
60 M Treated haemochromatosis, Transplant for HCC

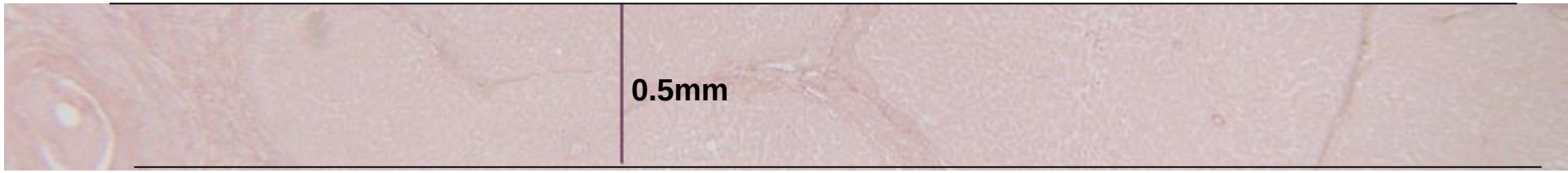

Van Gieson


Shikata’s Orcein

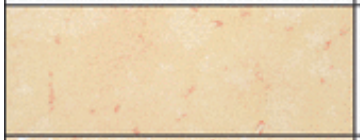
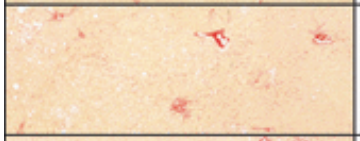
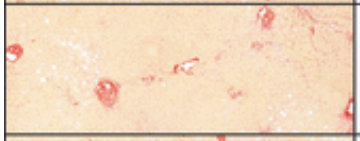
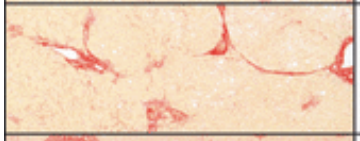
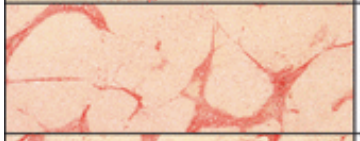
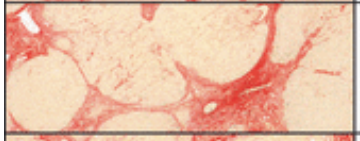
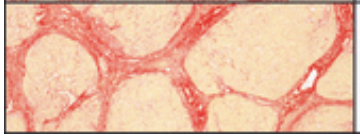

Shikata’s Orcein
64M hepatitis B cirrhosis


Shikata’s Orcein
1.0 mm


1.0 mm

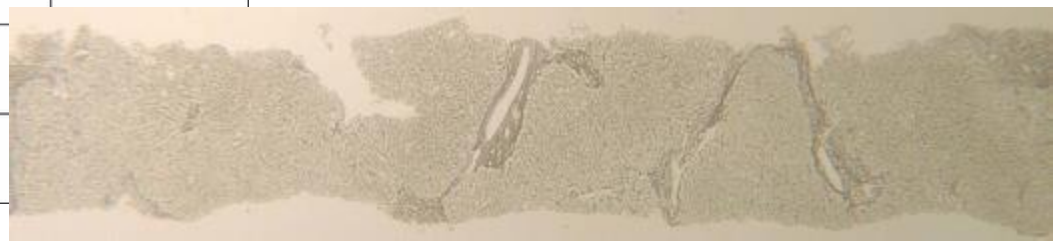

0.5mm

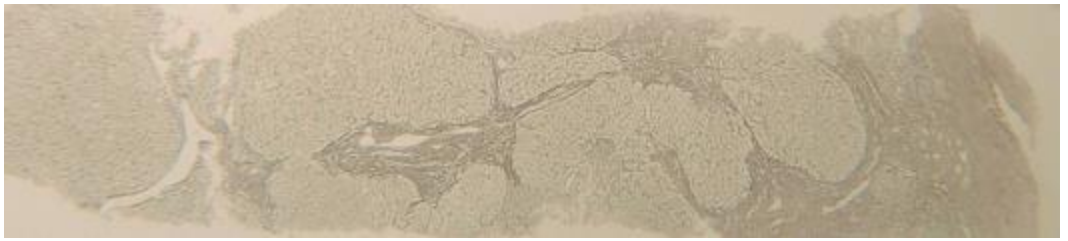
Chronic liver disease stage scores and quantitative liver fibrosis measurements.

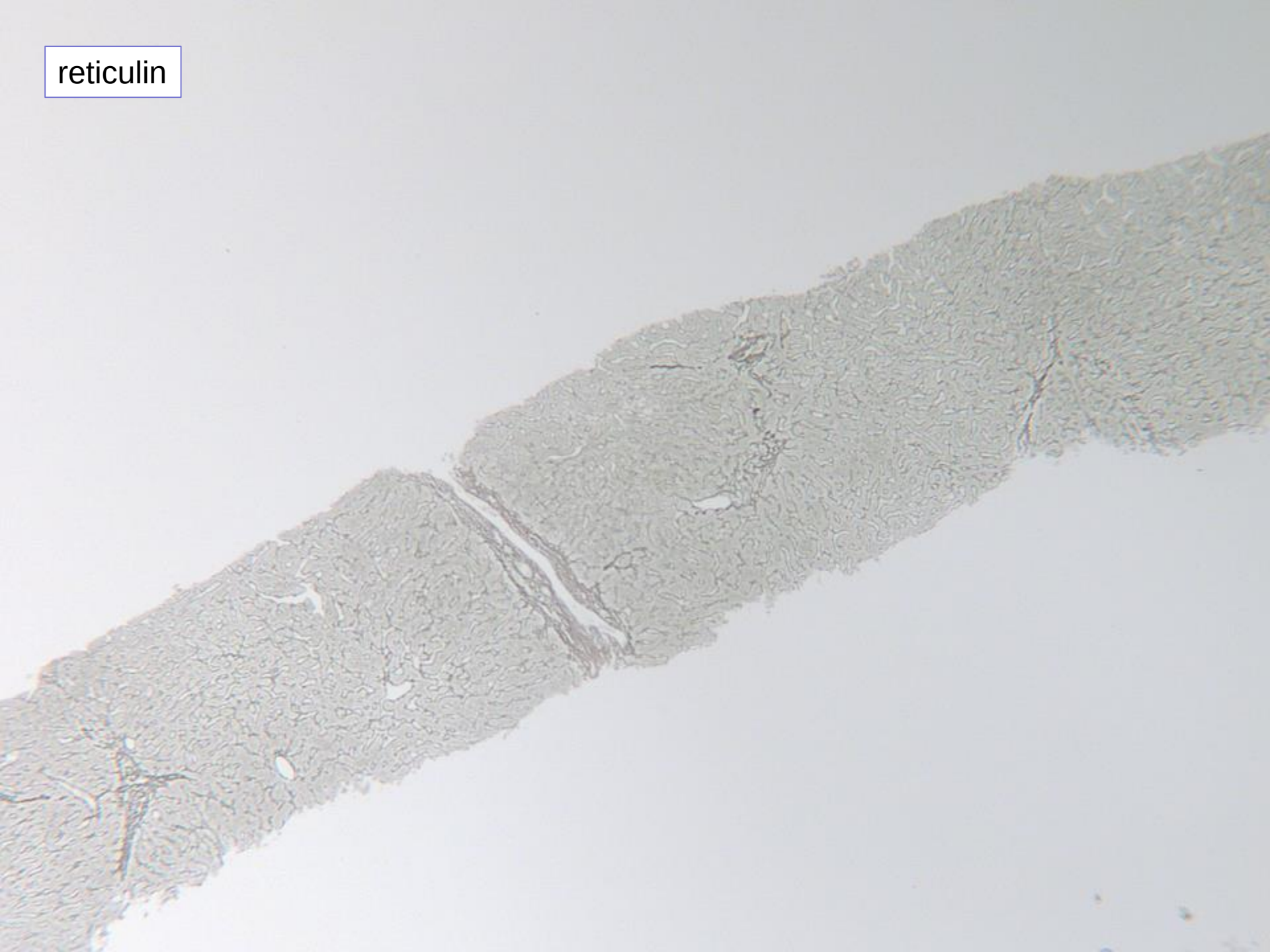
Appearance	Ishak stage: Categorical description	Ishak stage: Categorical assignment	Fibrosis measurement* (%)
	No fibrosis (normal)	0	1.9
	Fibrous expansion of some portal areas ± short fibrous septa	1	3.0
	Fibrous expansion of most portal areas ± short fibrous septa	2	3.6
	Fibrous expansion of most portal areas with occasional portal to portal (P-P) bridging	3	6.5
	Fibrous expansion of portal areas with marked bridging (portal to portal (P-P) as well as portal to central (P-C))	4	13.7
	Marked bridging (P-P and/or P-C), with occasional nodules (incomplete cirrhosis)	5	24.3
	Cirrhosis, probable or definite	6	27.8

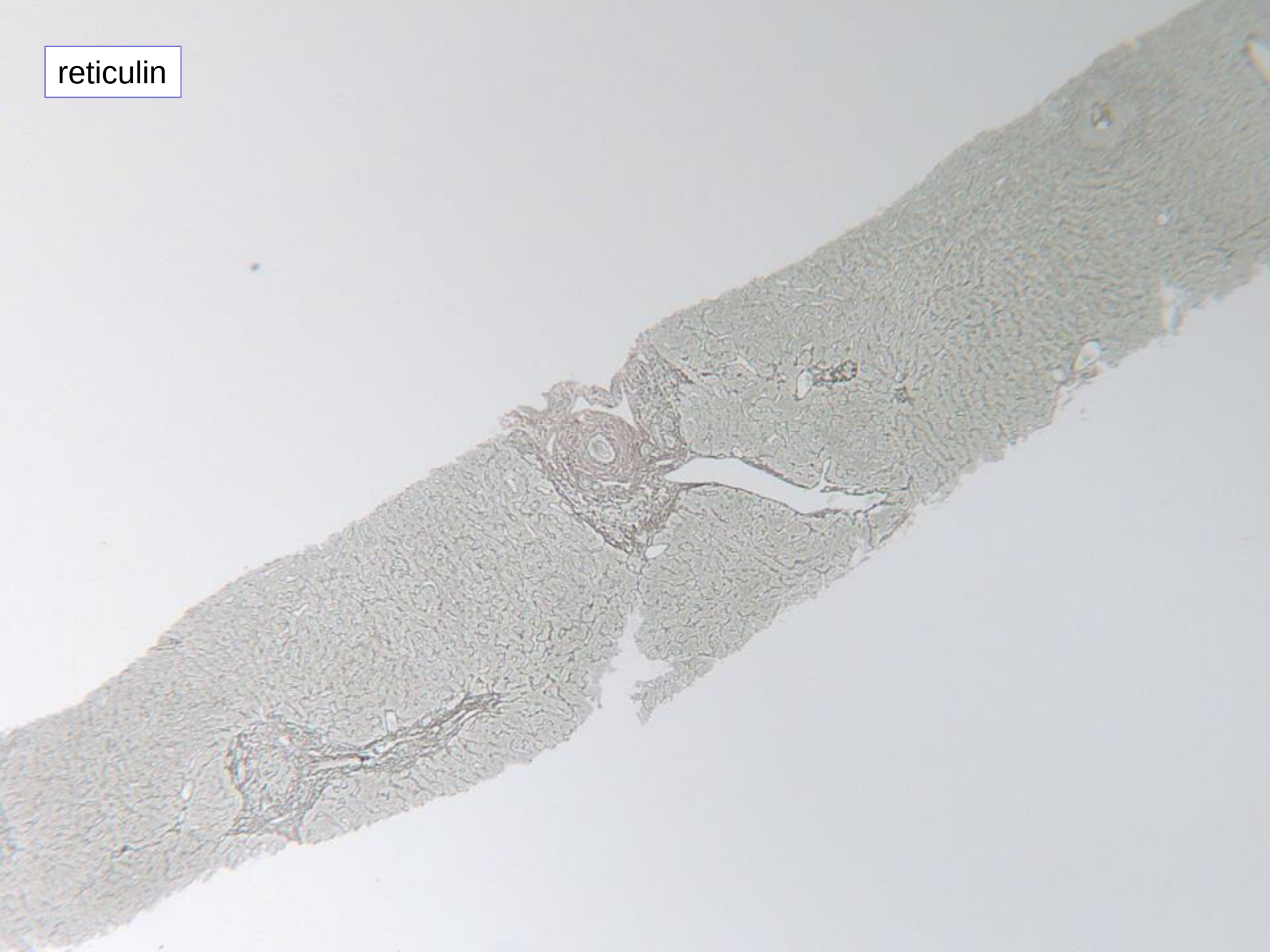
Standish RA *et al, Gut 2006;55;569-578*

Appearance	Ishak stage: Categorical description	Ishak stage: Categorical assignment	Fibrosis measurement* (%)
	No fibrosis (normal)	0	1.9
	Fibrous expansion of some portal areas ± short fibrous septa	1	3.0
	Fibrous expansion of most portal areas ± short fibrous septa	2	3.6
	Fibrous expansion of most portal areas with occasional portal to portal (P-P) bridging	3	
	Fibrous expansion of portal areas with marked bridging (portal to portal (P-P) as well as portal to central (P-C))	4	
	Marked bridging (P-P and/or P-C), with occasional nodules (incomplete cirrhosis)	5	
	Cirrhosis, probable or definite	6	

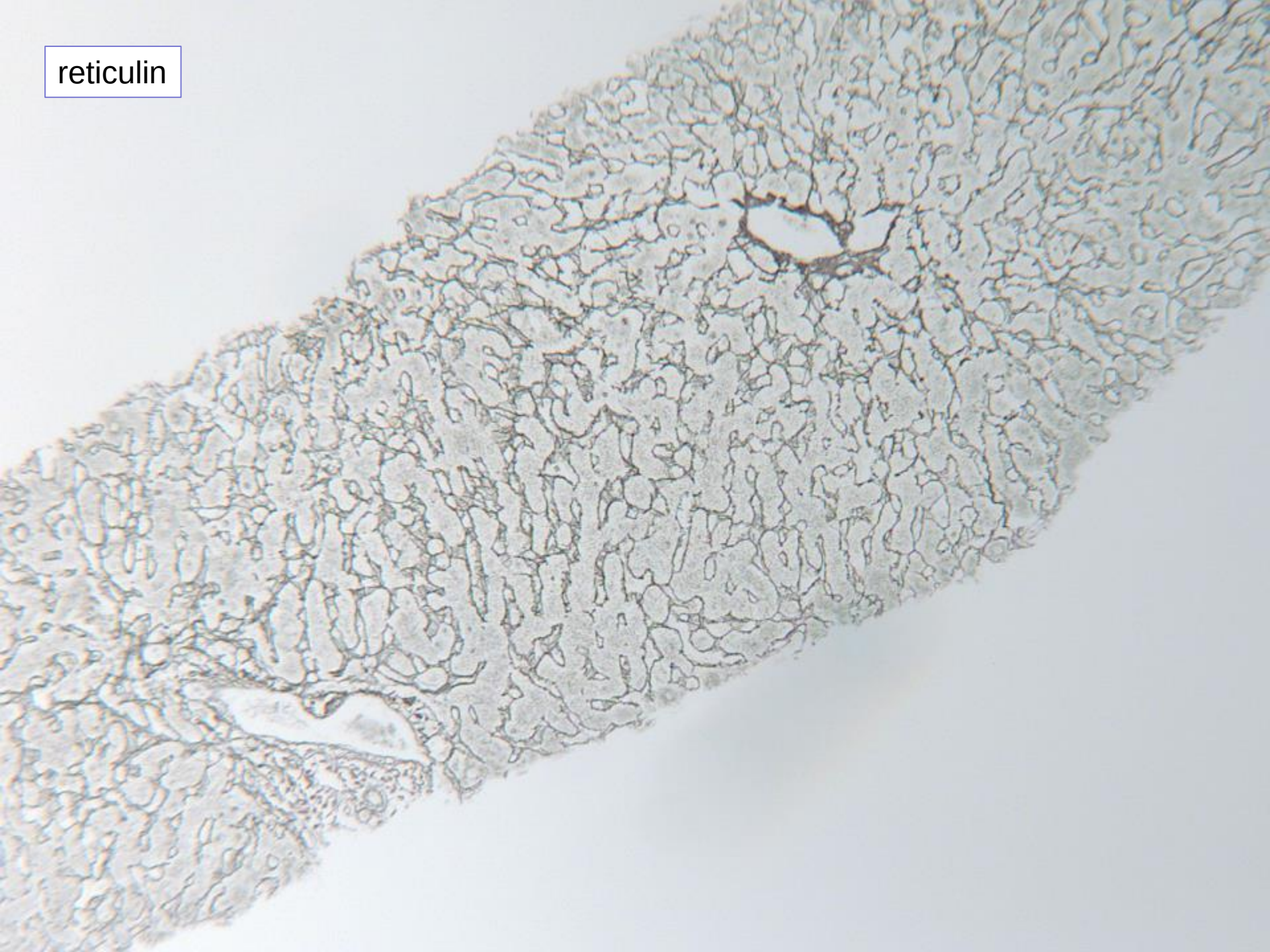


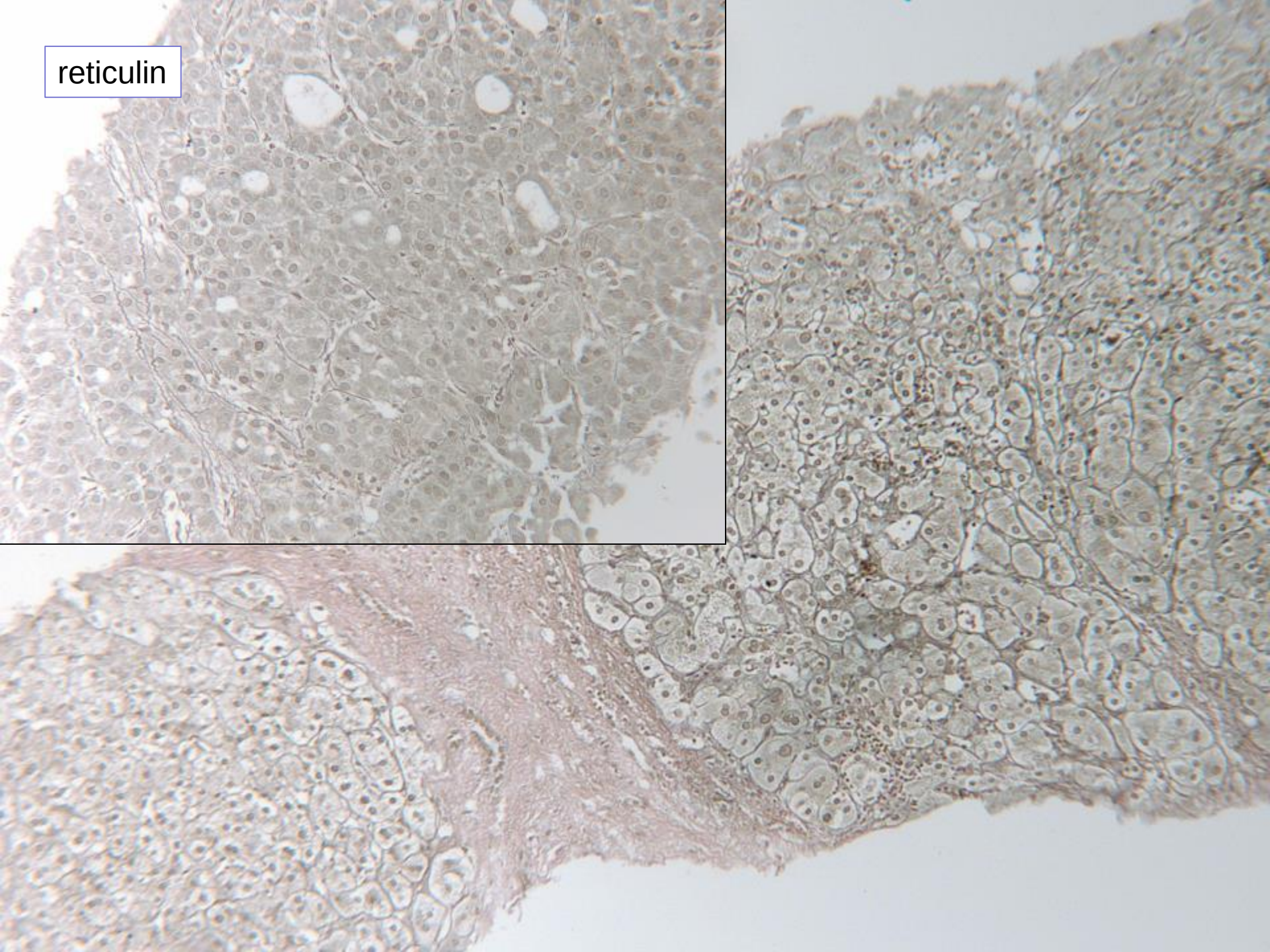



reticulin

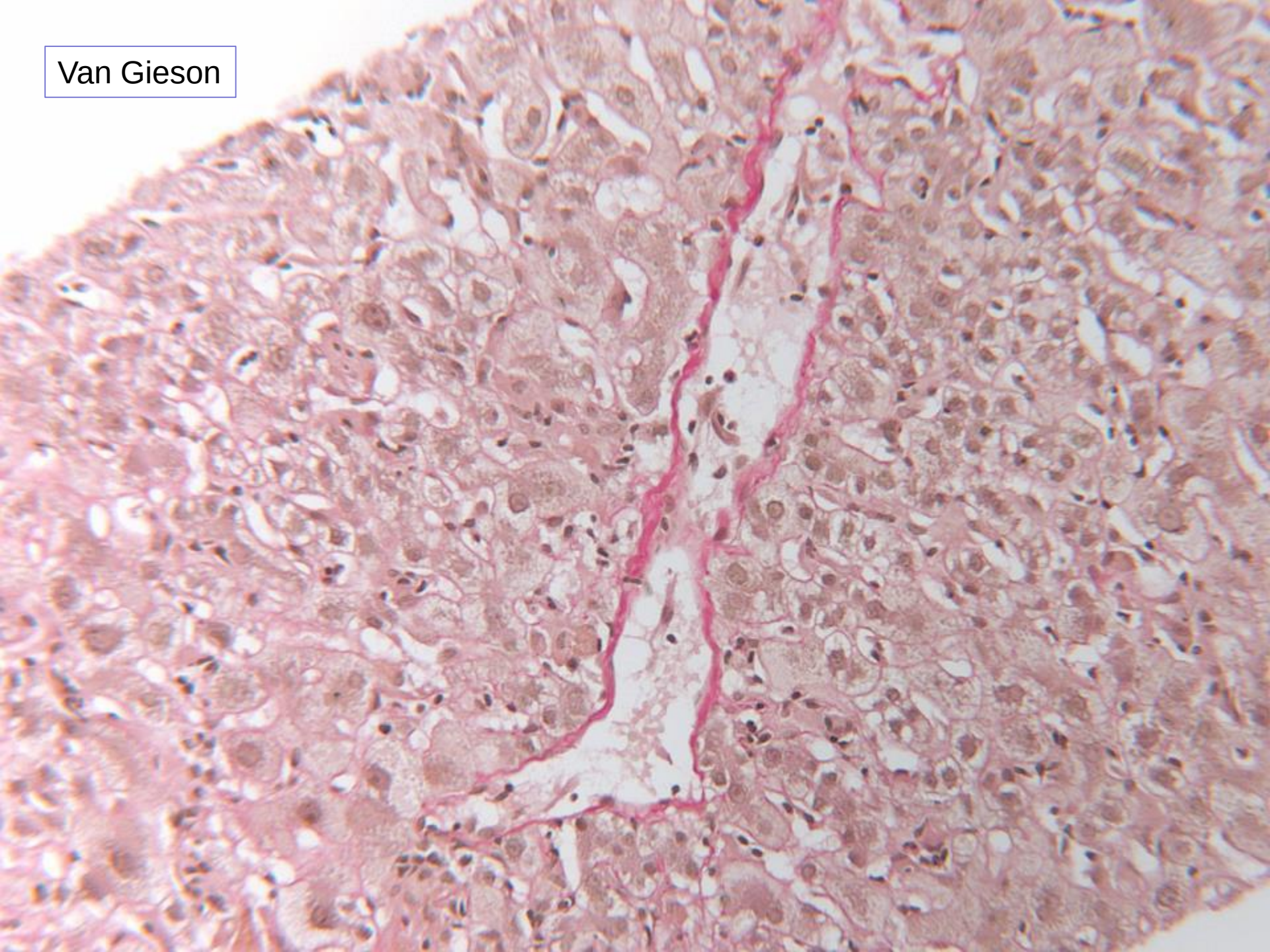

reticulin

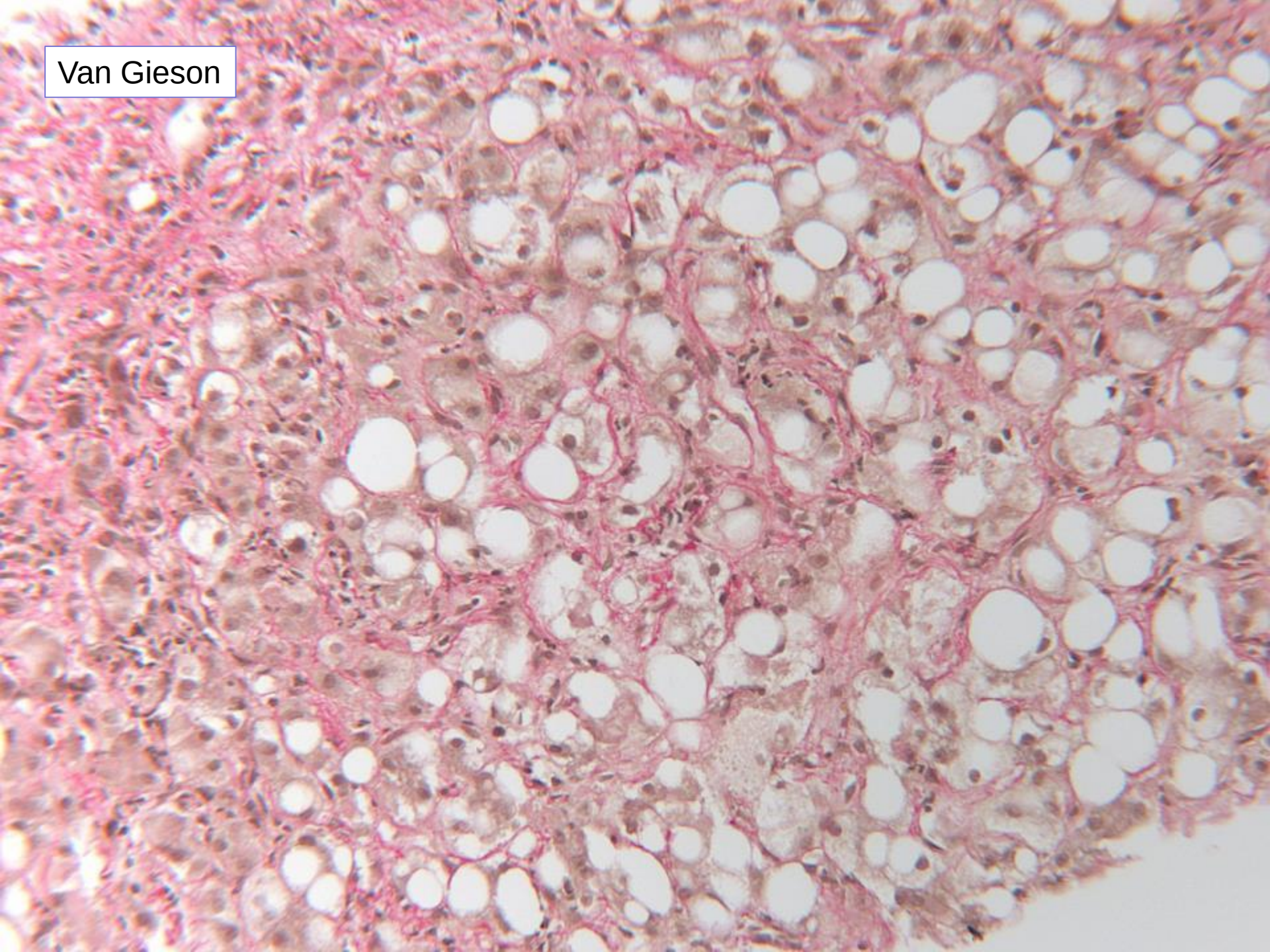
reticulin

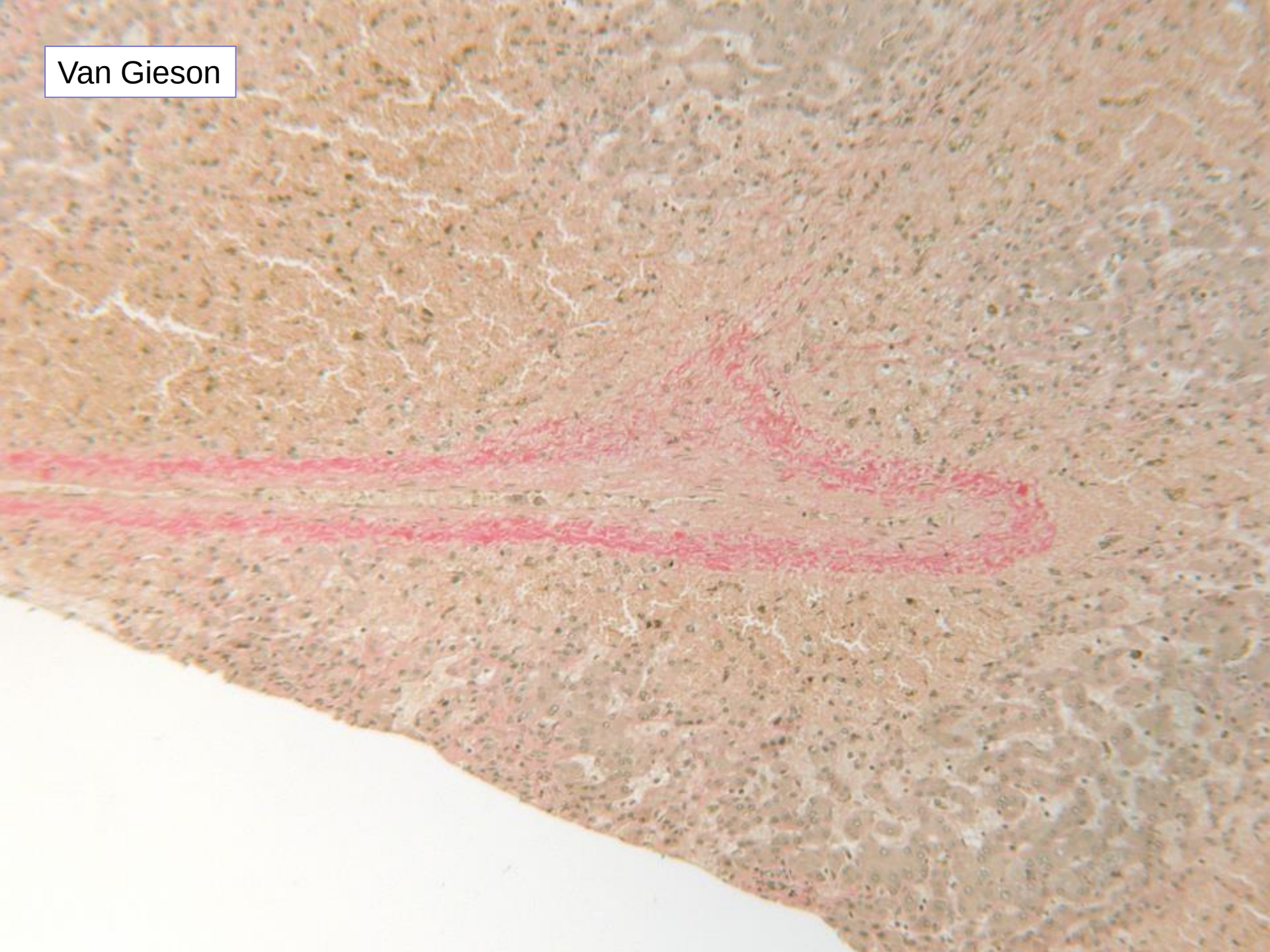



reticulin

reticulin


Van Gieson


Van Gieson


Van Gieson

Usual panel

Architecture:

- Retic
- van Gieson
- Shikata

Hepatocytes:

- PAS, PASD,
- Shikata

Pigment:

- Perl’s

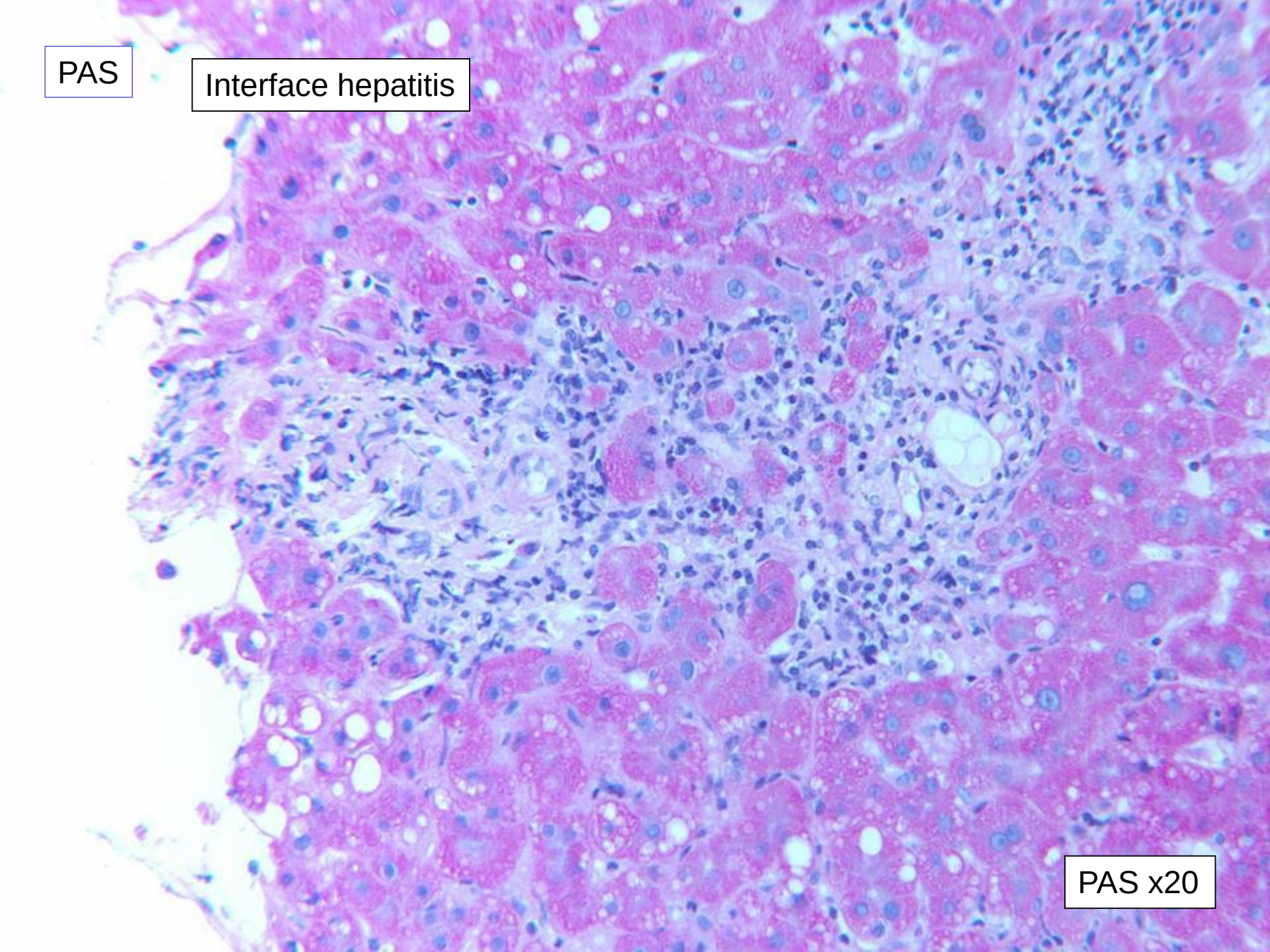
Usual panel

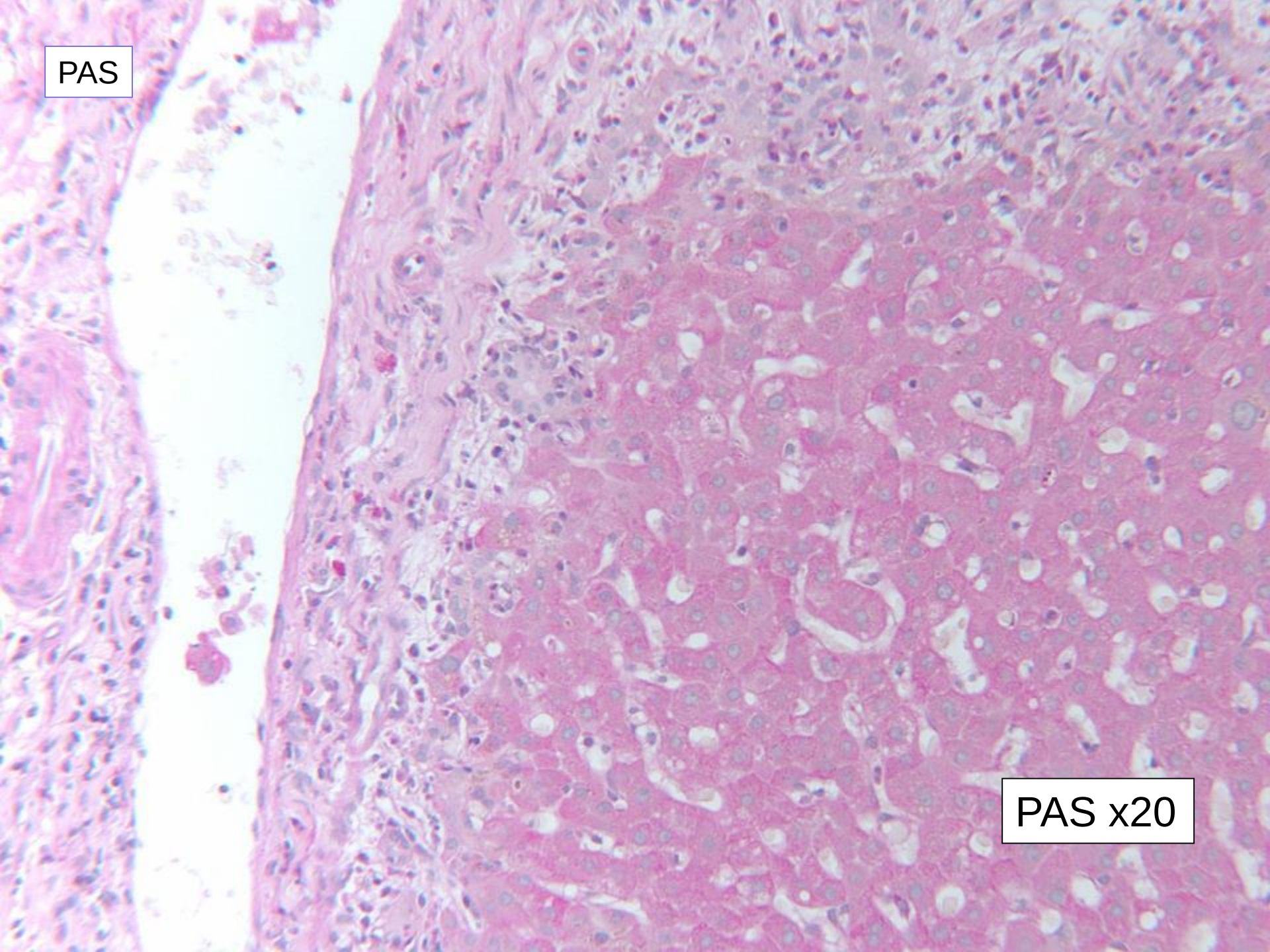
Architecture:
- Retic
- van Gieson
- Shikata

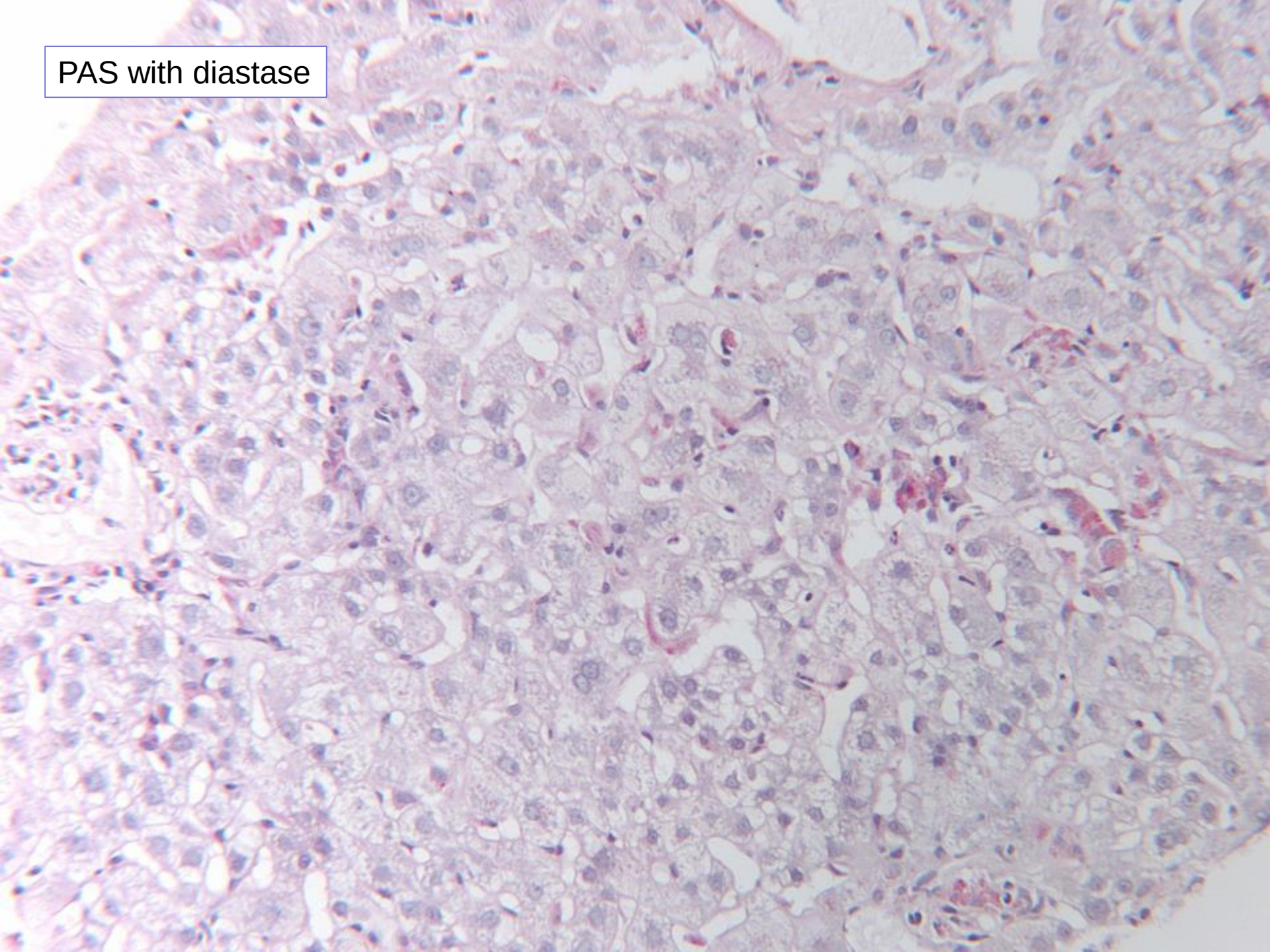
Hepatocytes:
- PAS – well fixed biopsies, demonstrates hepatocytes, limiting plate
- PASD – active Kupffer cells, A1ATD, bile ducts, basement membrane
- Shikata – copper-associated protein, HBsAg

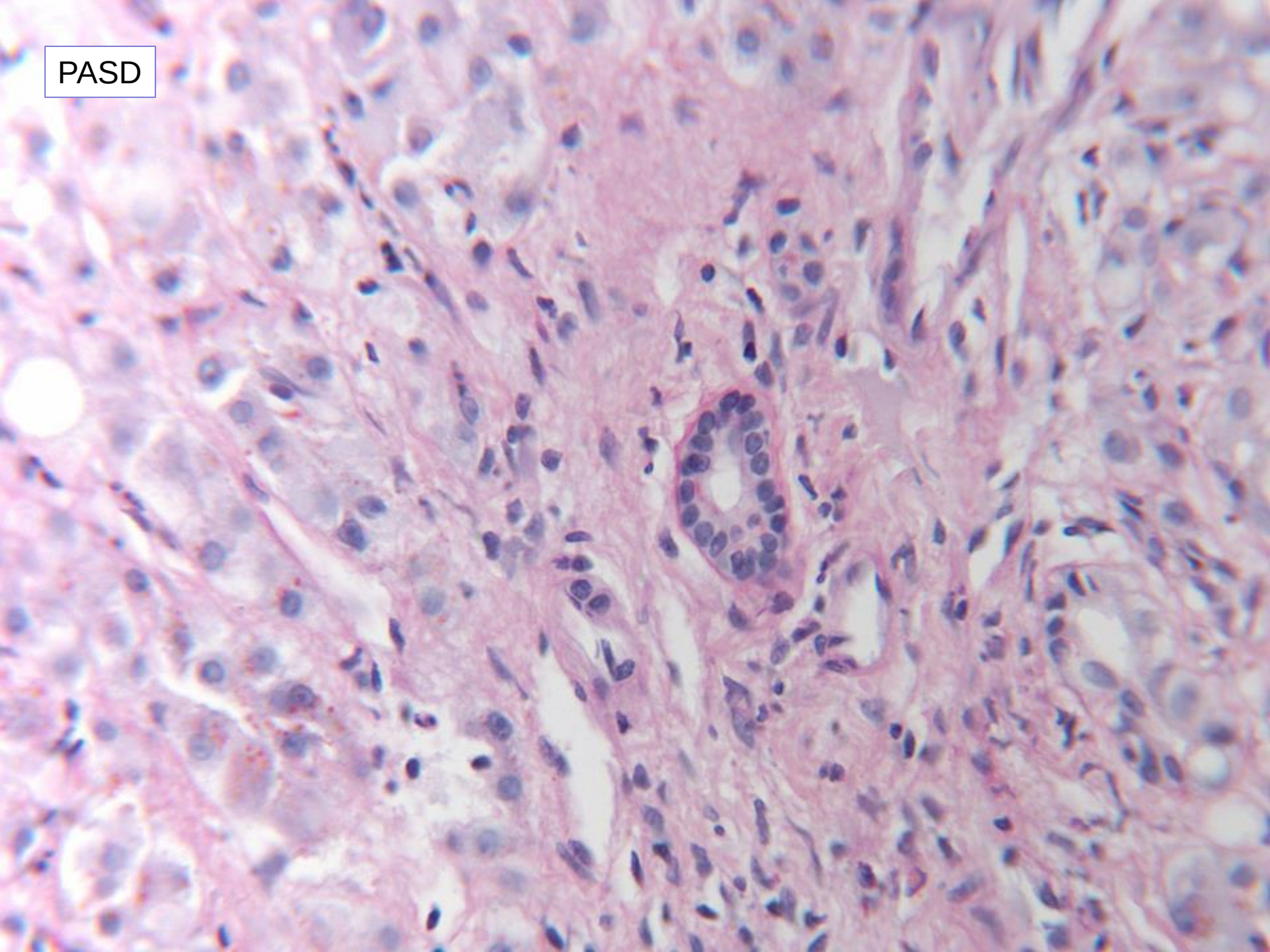
Pigment:
- Perl’s

Periodic Acid Schiff

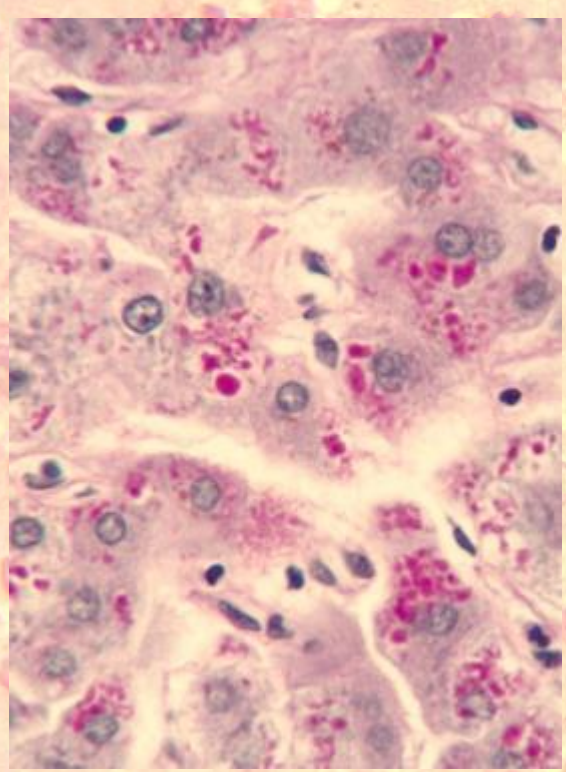

PAS
Interface hepatitis
PAS x20

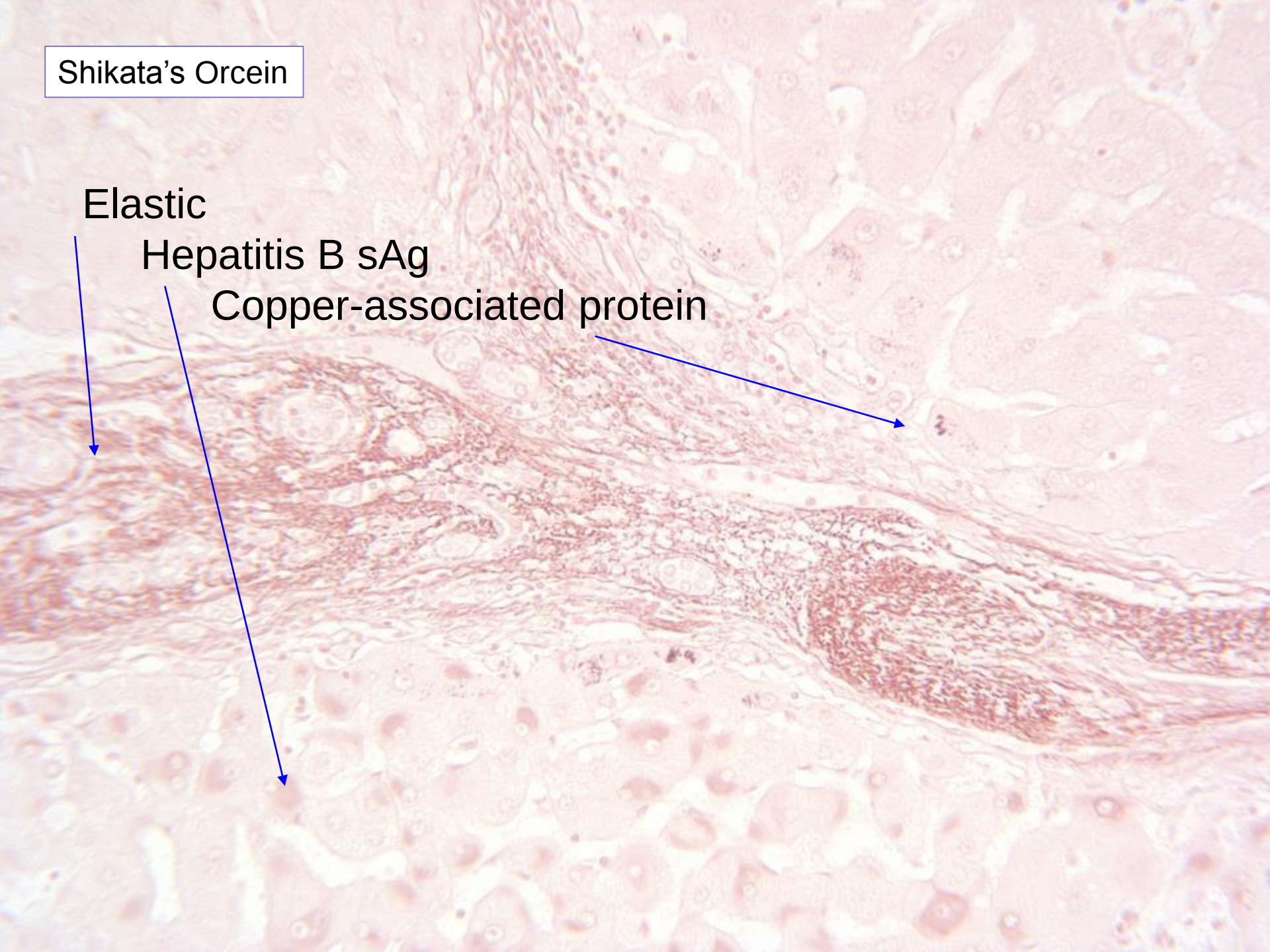

PAS
PAS x20

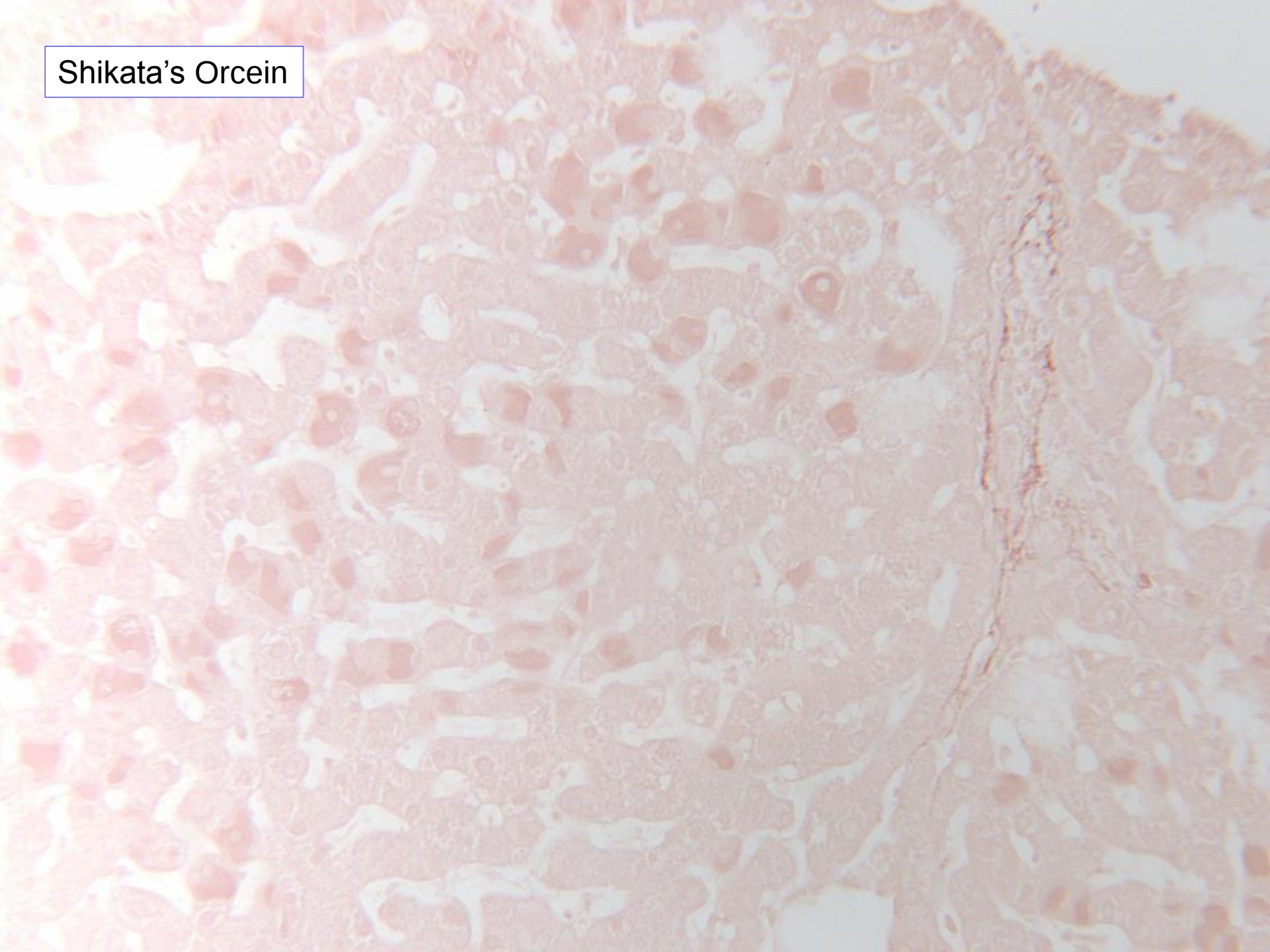

PAS with diastase

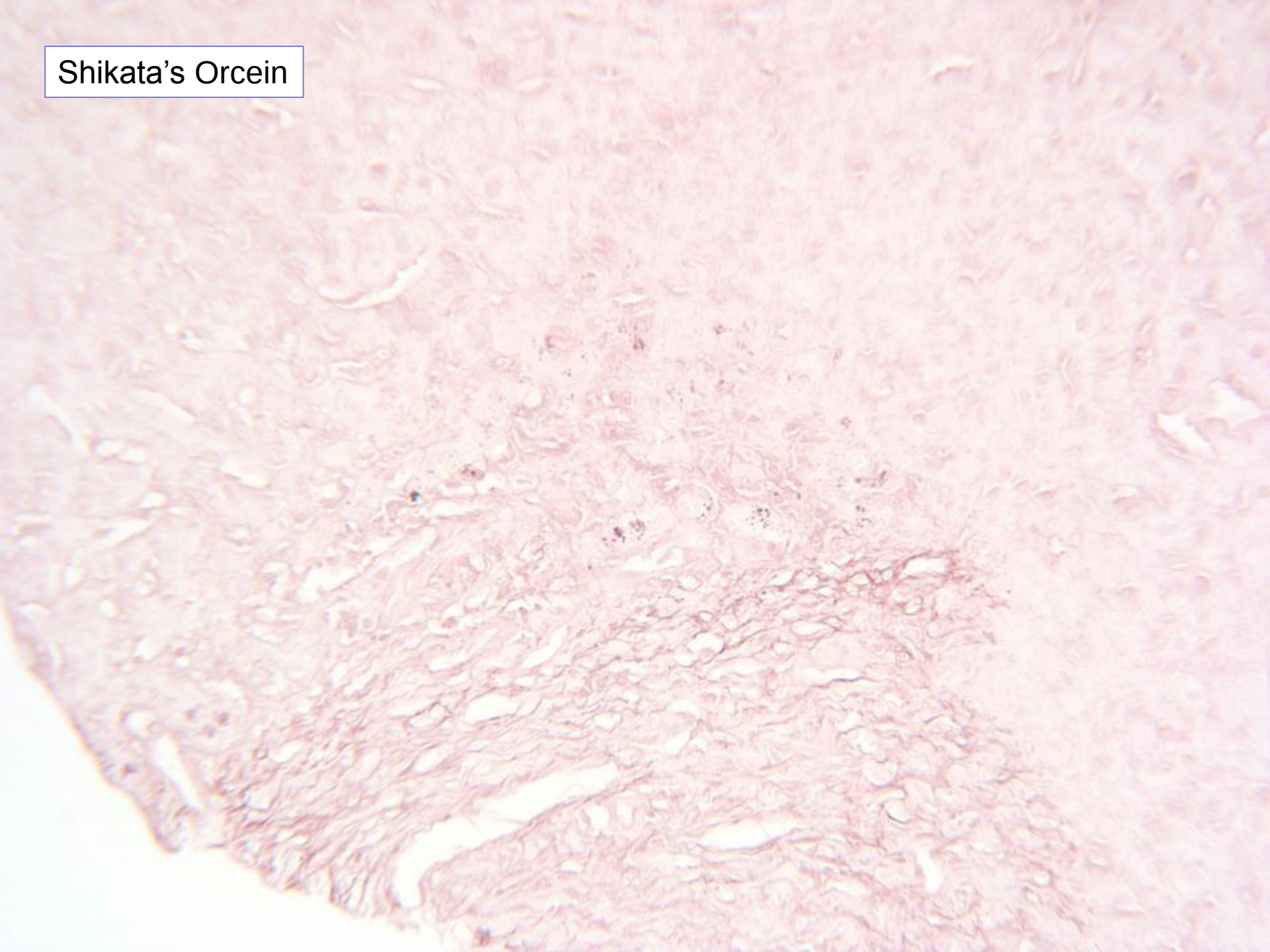

PASD

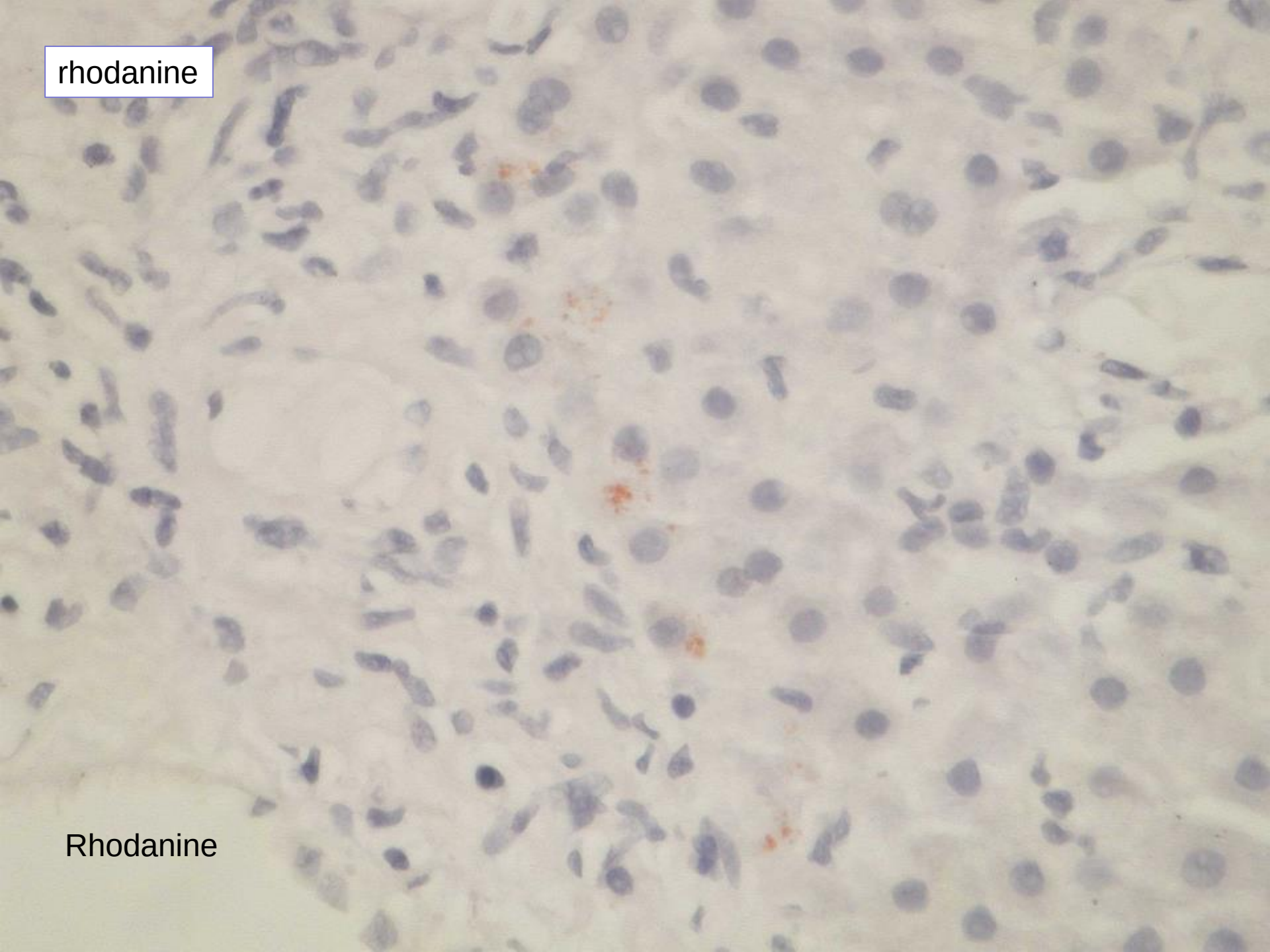
PASD



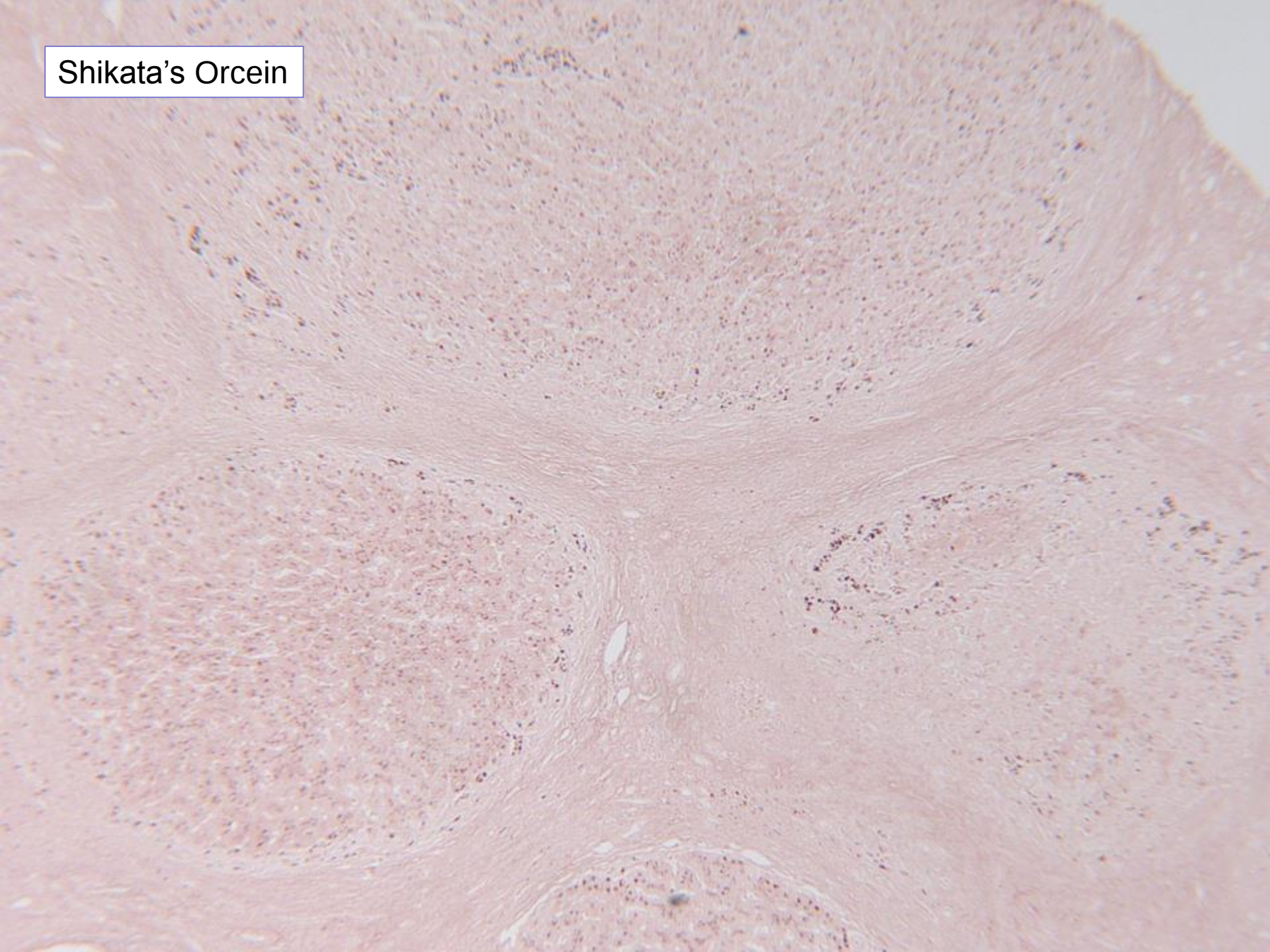

Shikata’s Orcein
Elastic
Hepatitis B sAg
Copper-associated protein


Shikata’s Orcein


Shikata’s Orcein



Rhodanine


Shikata’s Orcein

Usual panel

Architecture:

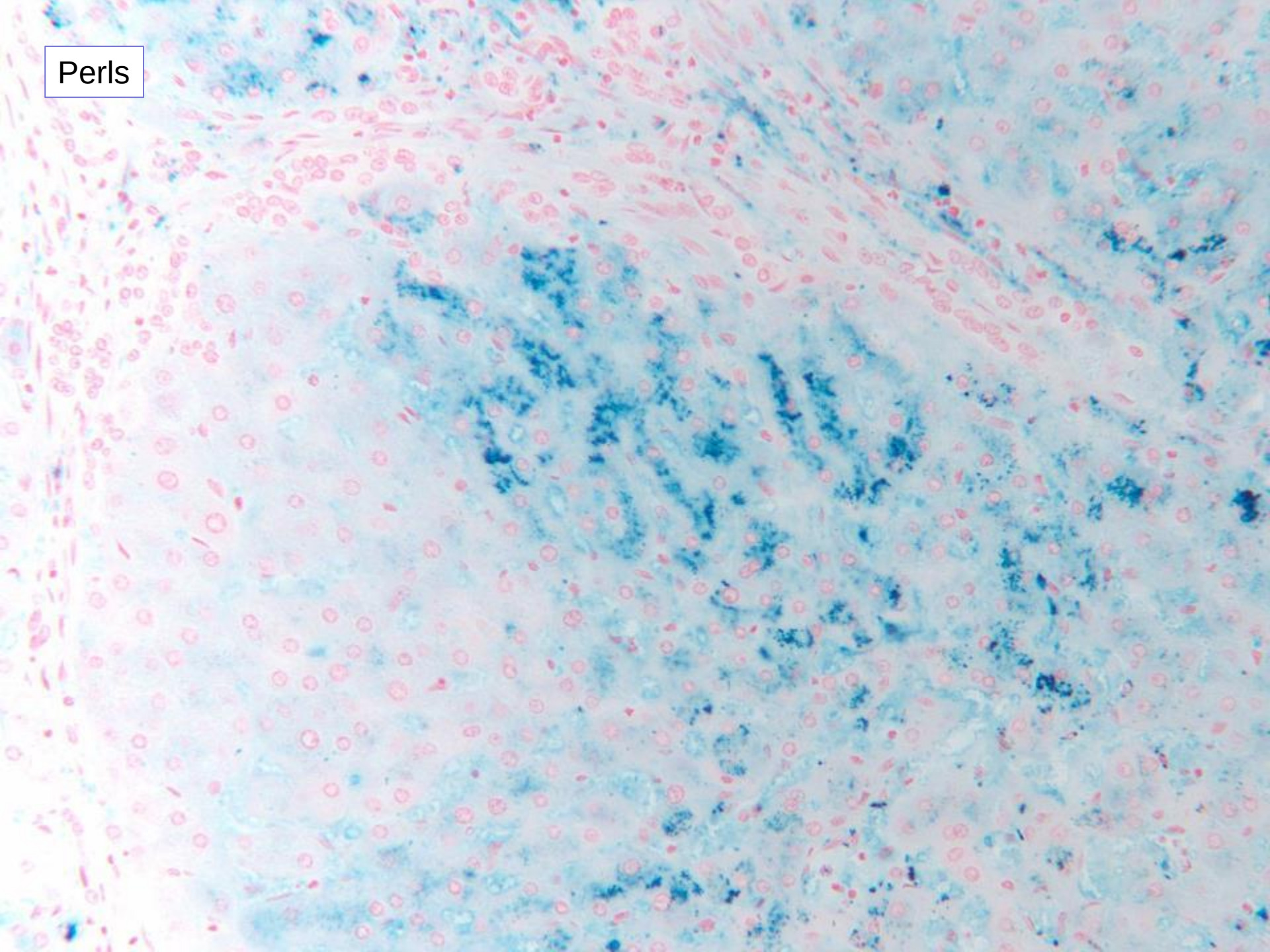
- Retic
- van Gieson
- Shikata

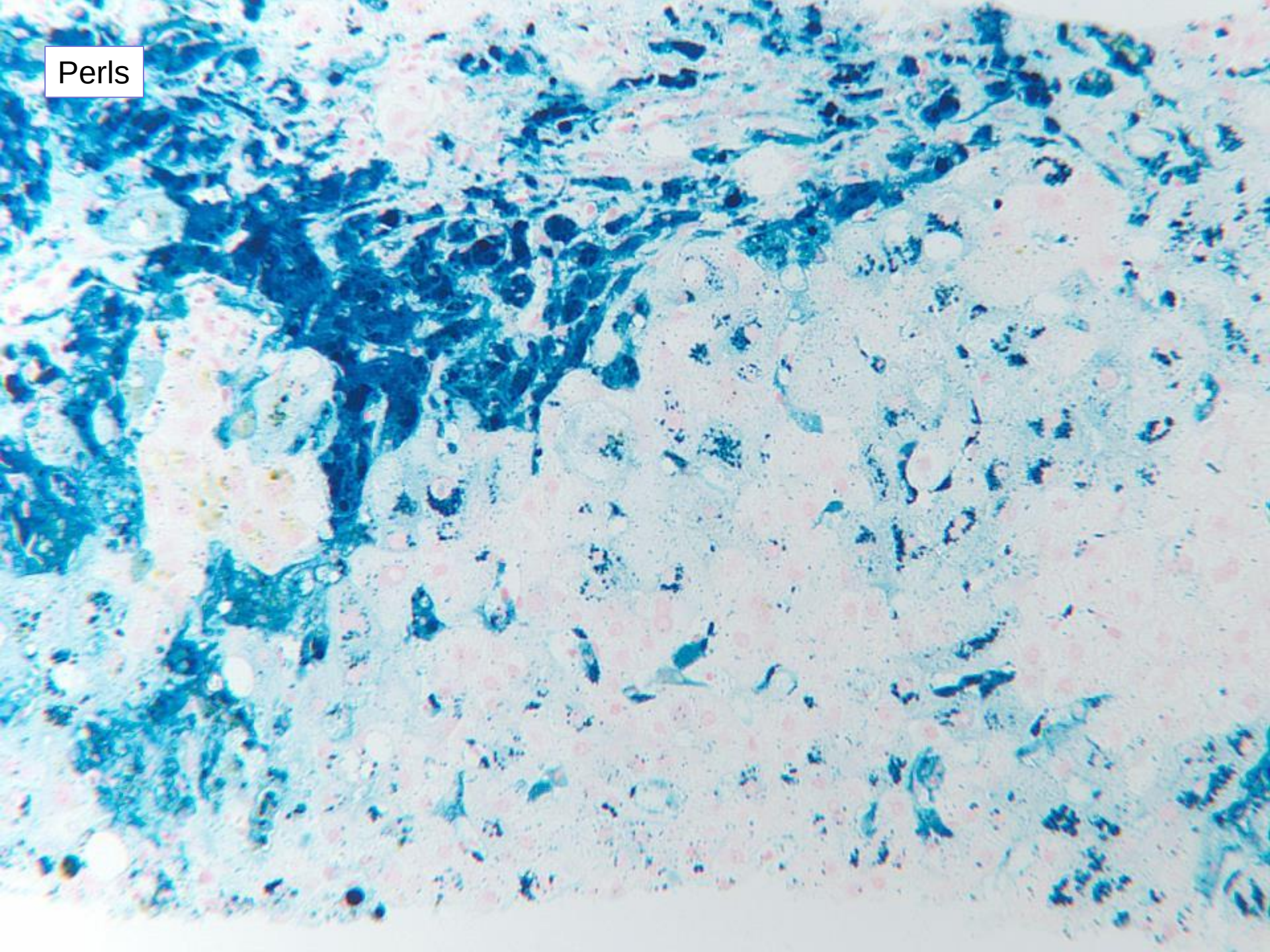
Hepatocytes:

- PAS, PASD,
- Shikata

Pigment:

- Perl’s – iron – hepatocytes, other cells


Perls


Perls

Iron in liver biopsies – distribution and cause

Predominant site of iron	Likely diagnosis
Hepatocytes only	haemochromatosis
Hepatocytes and Kupffer cells/endothelial cells	Transfusional overload haemolysis Increased absorption (alcohol) Late stage haemochromatosis
Kupffer cells	Anaemia of chronic disease Acute hepatitis

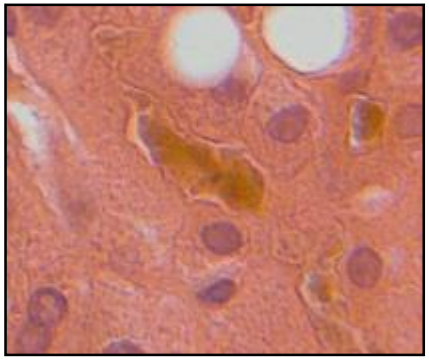
Pigments in liver biopsies

- Iron – periportal hepatocytes, sinusoidal cells

- Bilirubin – cytoplasmic, perivenular hepatocytes,
  also canaliculi, PASD +ve Kupffer cells
  occasionally bile ductules – suspect sepsis

- Ceroid / lipofuschin – cytoplasmic, perivenular hepatocytes
  can look very like bilirubin
  not canaliculi, not PASD+ve Kupffer cells
  if present = lack of recent hepatocyte injury

Bilirubinostasis – Why?? differential diagnosis



Look for additional features of:

a) **acute hepatitis** – lobular disarray with acidophil bodies, inflammatory cells

b) large bile duct obstruction – portal oedema, ductular reaction, ductular neutrophils

c) Sepsis – cholangiolitis with ductular bile plugs

d) **Drugs** – often mixed hepatitic/cholangiolitis

a) None of the above – bland cholestasis, drugs, **metabolic errors**

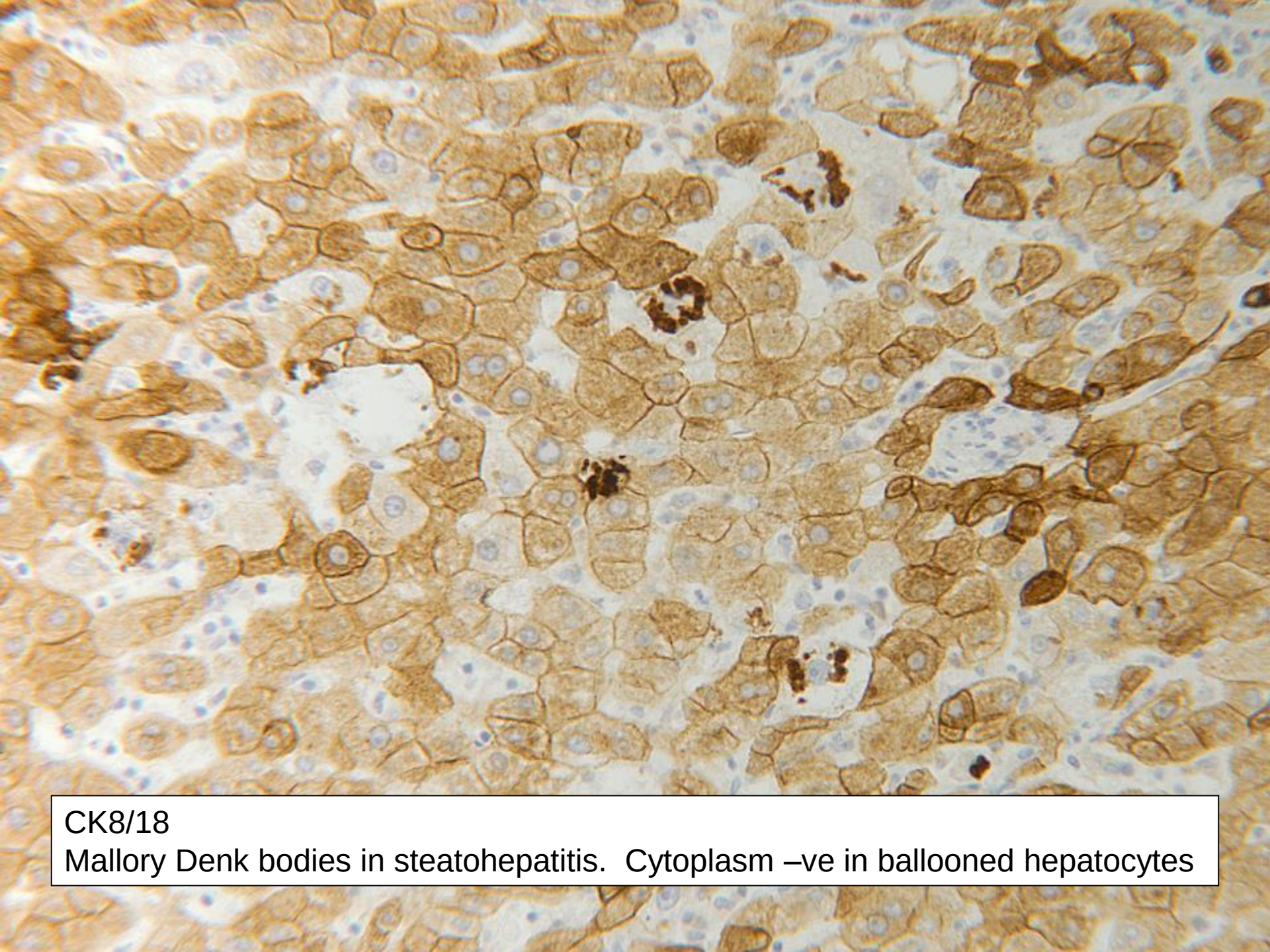
Immunohistochemistry?

- Mallory bodies in steatohepatitis
  - Ubiquitin
  - **CK8/18**
  - **Cam 5.2**

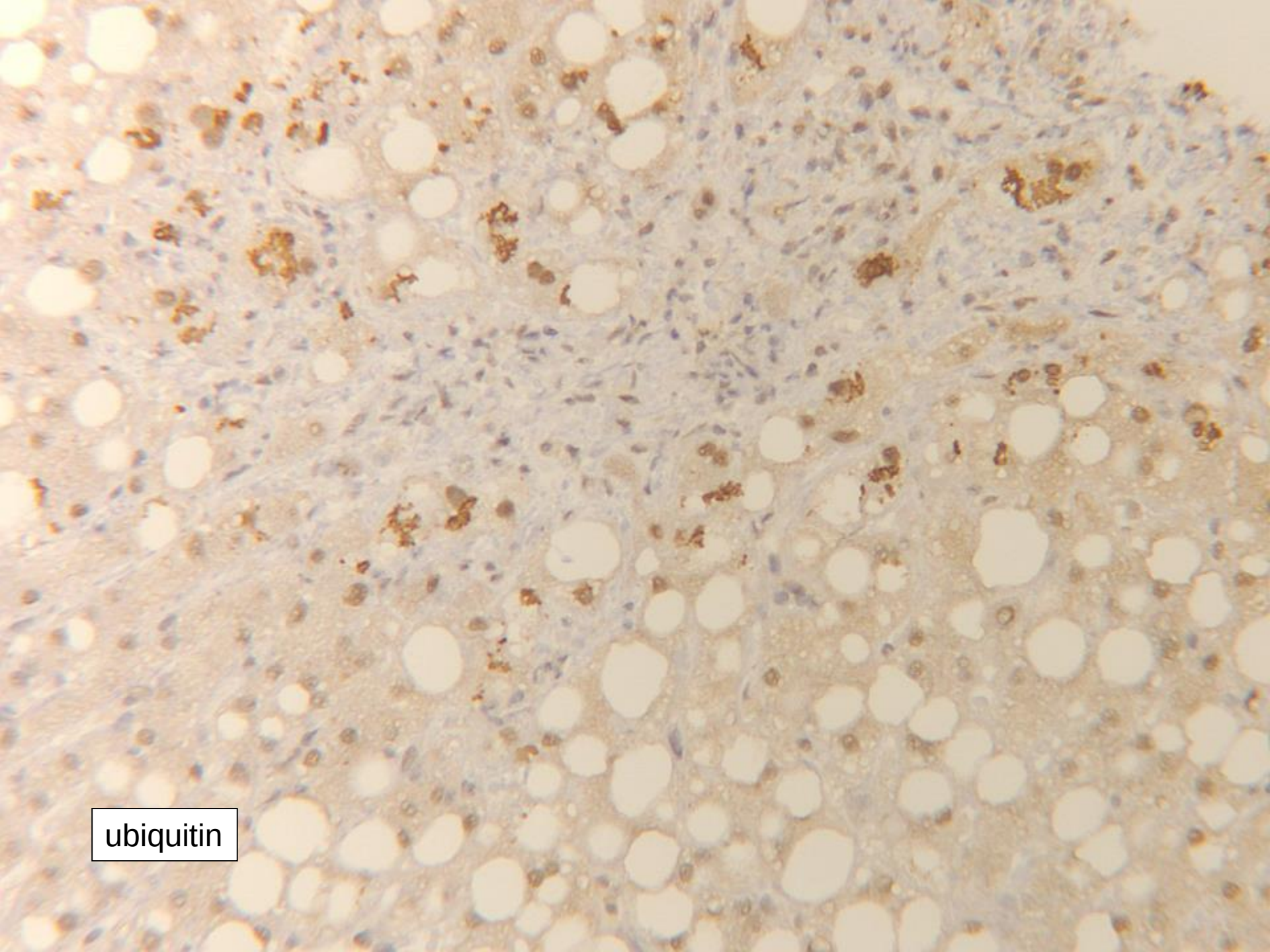
- Bile ducts and ductular reaction
  - CK7 – stains bile ducts
    and ‘intermediate hepatobiliary cells’ = +ve in hepatocytes

- Others – alpha 1 antitrypsin,
  viruses – hepatitis B, CMV, HSV

Tumours – another talk……..

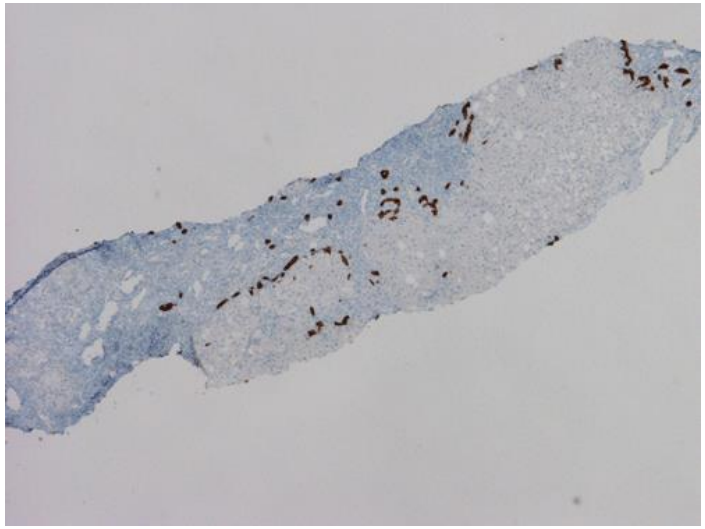


CK8/18
Mallory Denk bodies in steatohepatitis. Cytoplasm –ve in ballooned hepatocytes


ubiquitin

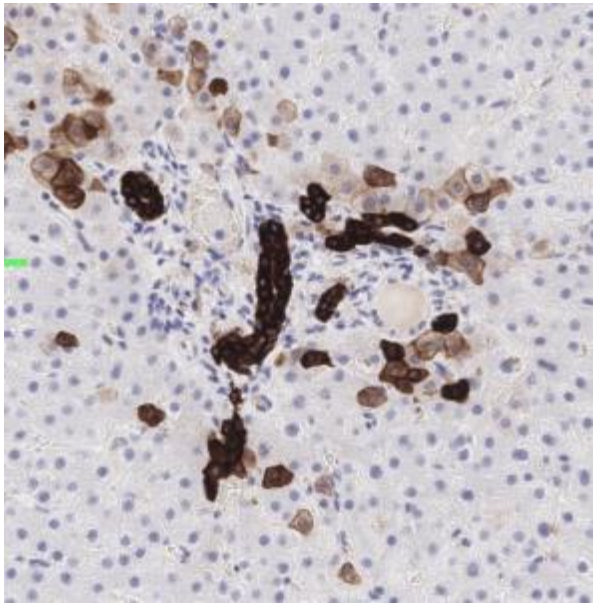
CK 7

in liver biopsies:

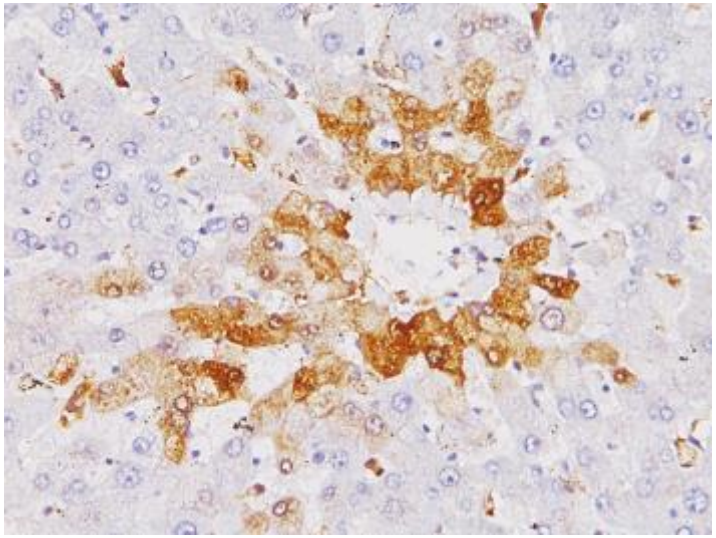


CK7+ in bile ducts and in ductules around fibrotic portal areas

Intermediate hepatobiliary cells



Periportal CK7+ hepatocytes in chronic biliary disease



CK7 +ve in
perivenular hepatocytes
- non-specific in chronic disease,

What is difficult?
Insights from comparison of original and review diagnosis in liver biopsies referred to Leeds Hepatology centre

Retrospective (140) and prospective (200),
similar discrepancy rate, 52%, half of these are 'major'

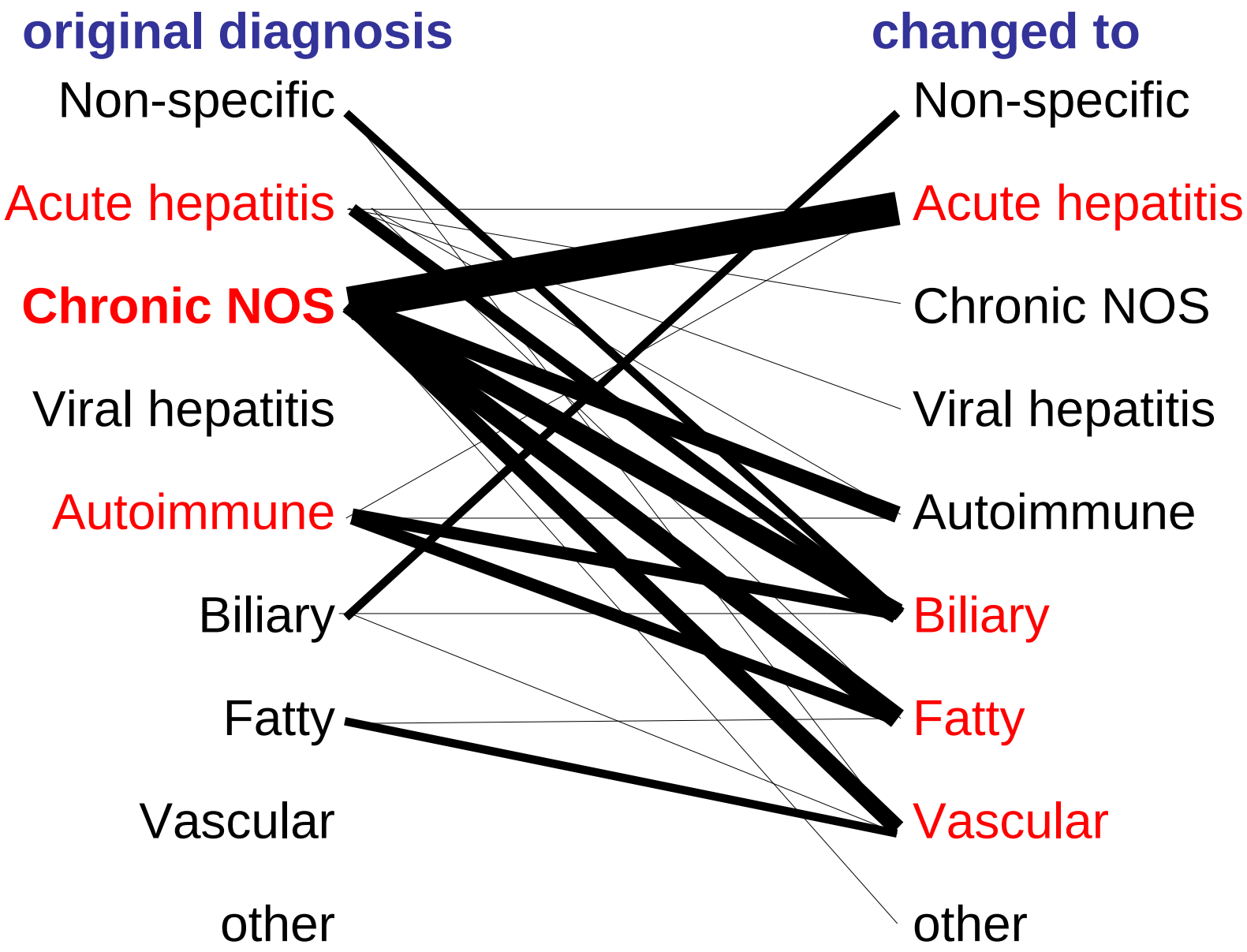
Final diagnoses most likely to have been missed were:

- Chronic biliary disease
- Acute hepatitis with bridging necrosis
  – interpreted as chronic liver disease
- Fatty liver disease – terminology and criteria
- Vascular diseases – rare and difficult to recognise

But for 'risk management' primary pathologist needs to know the pitfalls

– in which situations may important diagnoses be missed?

Major changes original v. revised diagnosis



Risk Management….

Chronic hepatitis: a morphological pattern, can you add the aetiology? needs clinicopathological correlation

Consider:

1) acute hepatitis with bridging necrosis v chronic hepatitis → • History, v. high ALT, Kupffer cells, VG collapse v. fibrosis

2) Exclude chronic biliary disease → • Shikata for copper-protein
High Alk Phos

3) Portal inflammation in steatohepatitis – increases while steatosis decreases → • Pericellular fibrosis, ballooning and Mallory bodies (CK8/18)
absence of cause of chronic hepatitis

4)Autoimmune hepatitis → • PCs, interface hepatitis, autoantibodies, IgG

5)Vascular → • Thin hepatocyte plates in areas of dilated sinusoids, sinusoidal fibrosis, inconspicuous portal veins

What to put in the report

- Clinical information
- Biopsy size / adequacy – length, portal tracts
- Architecture – vascular relationships, fibrosis, regeneration
- Main patterns of disease
- Special stains results
- Clinico-pathological comment
- Final concise bottom line & SNOMED

BRITISH LIVER TRUST

LOVE YOUR LIVER

- **There are many ways you can love your liver and help it function at its best, including:**
- **TAKE 2-3 DAYS IN A ROW OFF ALCOHOL**
- **STAY SLIM**
- **FEELING CONSTANTLY TIRED?**
- Go to your GP and ask for a liver function test.
- **DON'T LET IT GET TO THE STAGE WHERE YOU TURN YELLOW:**
- None of us want to look like Homer Simpson even if we may secretly admire some of his qualities. Many patients do end up looking like Homer (yellow and swollen) exactly because of those habits – beer, junk food and no exercise. Looking after your liver is not complicated and it's easy to prevent – it's just like looking after the rest of your body.