Autoimmune Hepatitis
The Role of Histopathology in Clinical Diagnosis and Management

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Role of Liver Biopsy in AIH

1. Establishing the diagnosis

2. Assessing disease severity
   - Inflammatory activity
   - Fibrosis

Role of Liver Biopsy in AIH

1. **Establishing the diagnosis**

2. Assessing disease severity
   - Inflammatory activity
   - Fibrosis

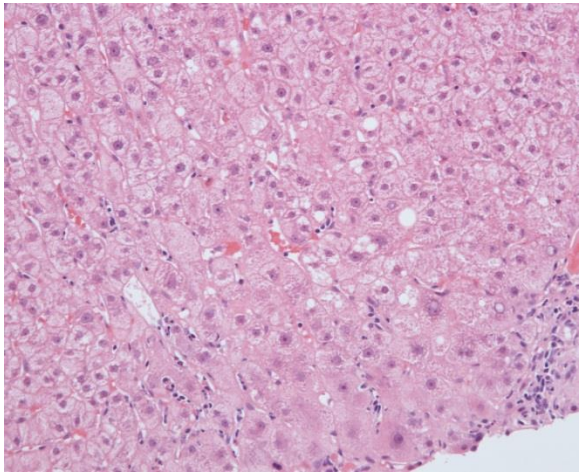
Autoimmune Hepatitis – Laboratory Investigations
Diagnostic Criteria

Biochemistry	Hepatitic LFTs • Raised AST/ALT
Immunology	Autoantibodies • ANA, SMA (type 1) • LKM , LC-1 (type 2) • SLA/LP (type 3) Immunoglobulins • Raised IgG
Histology	Presence of typical/compatible features Absence of atypical features (e.g. biliary features, fatty liver disease)

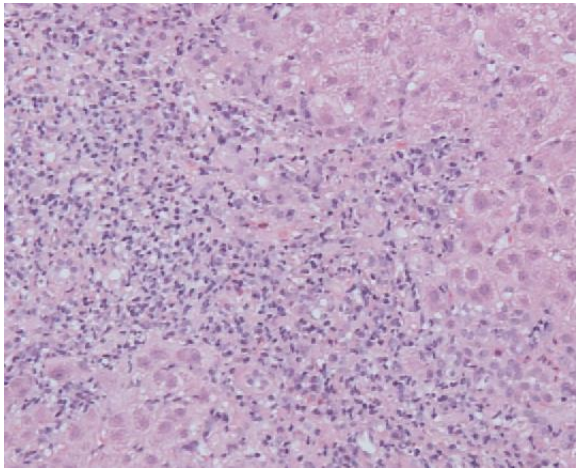
Liver Biopsy in AIH – Establishing the Diagnosis

Female, age 64

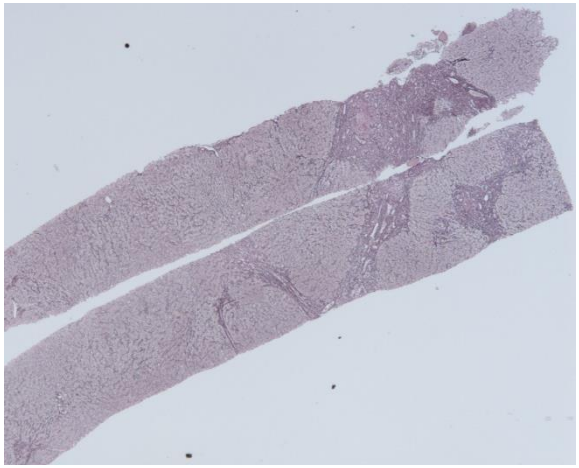
- Abnormal LFTs ? NAFLD, ? AIH.
- Initially suspected to have fatty liver disease (diabetes, hypertension, BMI 27).
- Subsequent investigations have revealed high titre antinuclear antibody and elevated serum IgG.



Minimal steatosis (<5%)



Portal lymphoplasmacytic inflammation with interface hepatitis



Bridging fibrosis (reticulin)

Conclusion

- Chronic hepatitis, in keeping with AIH.
- No significant fatty liver disease

International Autoimmune Hepatitis Group – Scoring Systems for Diagnosis of AIH

(Original –Johnson 1993, Revised – Alvarez 1999, Simplified – Hennes 2008)

- Various combinations of clinical, biochemical, immunological & histological features.
- Points allocated for typical features (deducted for atypical features in 1993 & 1999 systems).
- Total scores = “definite”, “probable” or “not” AIH.
- Original (1993) and Revised (1999) systems mainly intended for research purposes – e.g. clinical trials.
- Simplified (2008) system designed for routine practice.

Simplified Criteria for the Diagnosis of Autoimmune Hepatitis

Elke M. Hennes,[1] Mikio Zeniya,[2] Albert. J. Czaja,[3] Albert Parés,[4] George N. Dalekos,[5] Edward L. Krawitt,[6] Paulo L. Bittencourt,[7] Gilda Porta,[8] Kirsten M. Boberg,[9] Harald Hofer,[10] Francesco B. Bianchi,[11] Minoru Shibata,[12] Christoph Schramm,[1] Barbara Eisenmann de Torres,[13] Peter R. Galle,[13] Ian McFarlane,[14] Hans-Peter Dienes,[15] Ansgar W. Lohse,[1] and the International Autoimmune Hepatitis Group

(HEPATOLOGY 2008;48:169-176.)

Table 2. Simplified Diagnostic Criteria for Autoimmune Hepatitis

Variable	Cutoff	Points
ANA or SMA	≥1:40	1
ANA or SMA	≥1:80	
or LKM	≥1:40	2*
or SLA	Positive	
IgG	>Upper normal limit	1
	>1.10 times upper normal limit	2
Liver histology (evidence of hepatitis is a necessary condition)	Compatible with AIH	1
	Typical AIH	2
Absence of viral hepatitis	Yes	2

≥6: probable AIH
≥7: definite AIH

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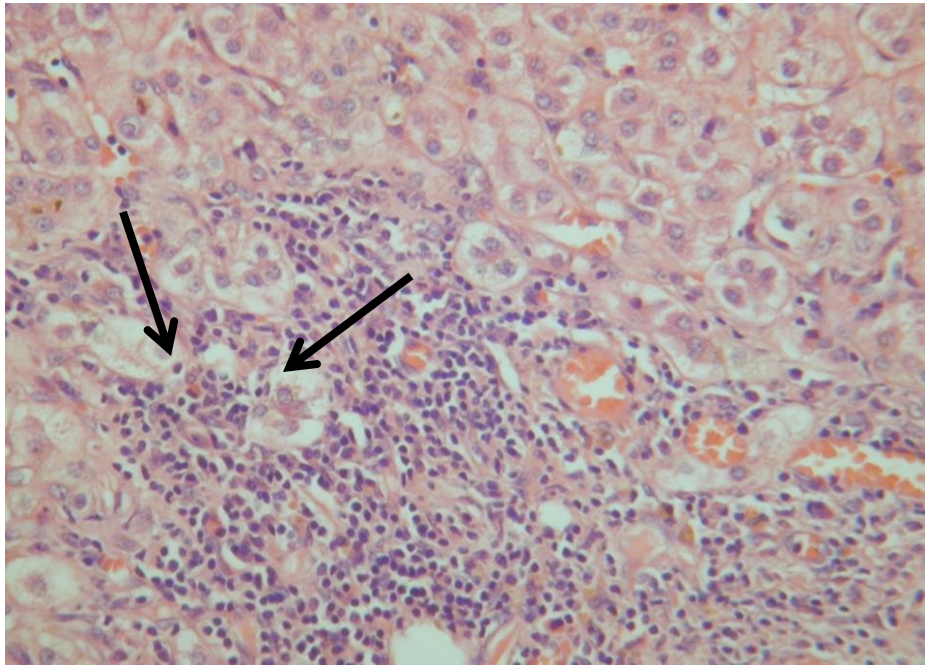
Criteria for Classifying Histology

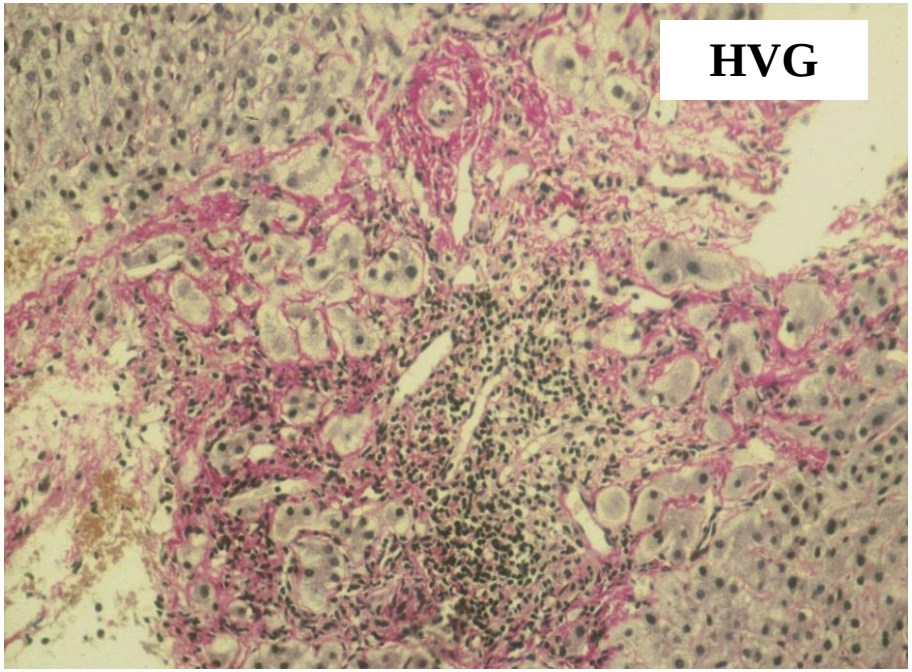
Typical of AIH (Score = 2 points)	Portal lymphoplasmacytic infiltration with **all three of**: 1. Interface hepatitis 2. Hepatocyte rosettes 3. Lymphocyte emperipolesis
Compatible with AIH (Score = 1 point)	Portal lymphoplasmacytic infiltration without one or more of the three "typical features"
Atypical for AIH (Score = 0 points)	Features suggest another diagnosis – e.g. steatohepatitis

Problems / Limitations

1. Scoring system designed for patients presenting with chronic hepatitis.
2. Assessment of hepatocyte rosettes and emperipolesis.

Chronic Autoimmune Hepatitis -Typical Features





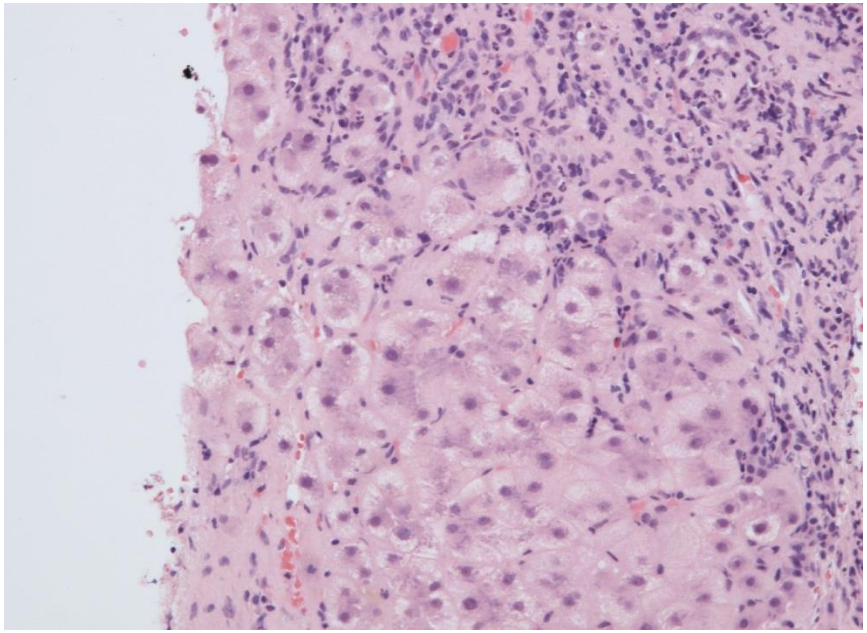
Interface Hepatitis

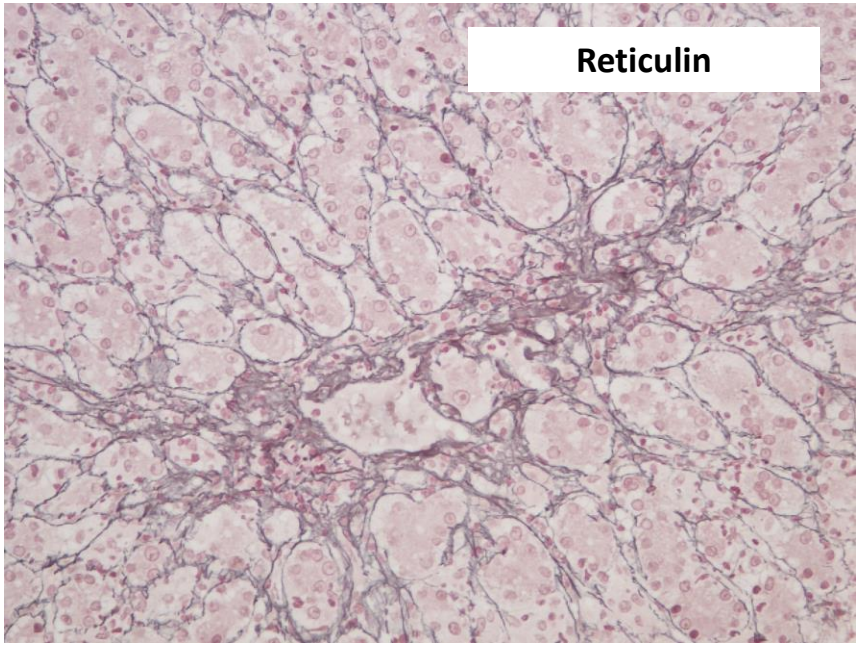
- Often associated with ballooning and rosetting of periportal hepatocytes
- May also be associated with emperipolesis of lymphocytes in periportal hepatocytes

Periportal Fibrosis

- Delicate periportal fibrous strands often surrounding hepatocyte rosettes
- Connective tissue stains (e.g. HVG/Trichrome) required to identify early periportal fibrosis

Hepatocyte Rosettes





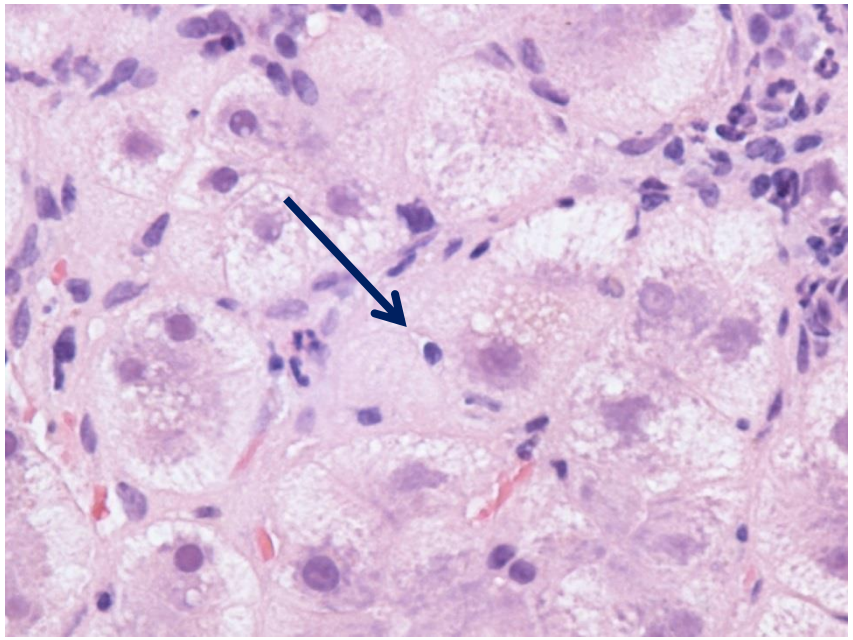
Hepatitic Rosettes

- Clusters of hepatocytes embedded in connective tissue
- May be a manifestation of regenerative activity with hyperplastic hepatocytes trapped in areas of inflamed/collapsed liver parenchyma
- Present in areas of moderate/severe interface hepatitis in cases of chronic (portal) hepatitis – e.g. autoimmune hepatitis
- Also frequently present in cases of acute (lobular) hepatitis

Emperipolesis

Definition

Presence of an intact (viable) cell within the cytoplasm of another cell



Relevance to Liver Disease

- Engulfment of lymphocytes (or other inflammatory cells) by hepatocytes, probably reflecting immune-mediated injury
- Occurs in several liver diseases (including AIH, HBV, HCV)
- May be important in disease pathogenesis

- Can be difficult to identify (e.g. distinction from hepatocyte nuclei and sinusoidal lymphocytes)

What is the utility of the Simplified Scoring System in distinguishing AIH from other inflammatory liver diseases histologically?

Assessment of the Histopathological Key Features in Autoimmune Hepatitis

De Boer, Histopathology 2015; 66: 351 - 362

Detailed assessment (blinded to clinical data) of presence/severity of relevant histological features:

- **63 pre-treatment biopsies from AIH patients**
- **62 biopsies from patients with untreated chronic viral hepatitis (41 HCV, 21 HBV)**

Table 3. Histological features in autoimmune hepatitis (AIH) and chronic viral hepatitis

Histological features	AIH, *n* (%)	Viral hepatitis, *n* (%)	*P*-value	Odds ratio	95% Confidence interval
Portal–periportal activity (≥2)*	55 (87)	39 (63)	0.002	4.1	1.6–10.0
Lobular activity (=4)*	25 (40)	4 (7)	<0.001	9.5	3.1–29.6
Fibrosis (=4)*†	4 (12)	7 (12)	1.0	1.0	0.3–3.6
Plasma cells (≥2)	30 (48)	17 (27)	0.02	2.4	1.1–5.1
Rosettes (≥2)‡	23 (49)	13 (23)	0.004	3.4	1.5–8.1
Emperipolesis (≥2)	49 (78)	31 (50)	0.001	3.5	1.6–7.6
Any biliary inflammation (≥1)	44 (70)	24 (39)	<0.001	3.6	1.7–7.7
Moderate–severe cholangitis (≥2)	18 (29)	11 (18)	0.2	1.9	0.8–4.3
Steatosis (≥2)	2 (3)	9 (15)	0.03	0.2	0.04–0.9
Ductopenia	4 (6)	2 (3)	0.7	2.0	0.4–11.5
Periductal concentric fibrosis	2 (3)	0 (0)	0.5	–	–
Endophlebitis	9 (15)	1 (2)	0.008	10.6	1.3–86.1
Granulomas	7 (11)	1 (2)	0.06	7.6	0.9–63.9

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1 → Portal–periportal activity (≥2)*

2 → Rosettes (≥2)‡

3 → Emperipolesis (≥2)

1,2 & 3 = "Typical Features" for AIH (Hennes 2008)

Assessment of the Histopathological Key Features in Autoimmune Hepatitis

De Boer, Histopathology 2015; 66: 351 - 362

Table 4. Sensitivity and specificity of 'typical' autoimmune hepatitis features in autoimmune hepatitis and chronic viral hepatitis

Features	Sensitivity (%)	Specificity (%)
Interface hepatitis (≥2)	84	36
Plasma cells (≥2)	49	71
Rosettes (≥2)	51	77
Emperipolesis	78	50
Interface hepatitis + rosettes + plasma cells ('criteria 1999')	27	91
Interface hepatitis + rosettes + emperipolesis ('criteria 2008')	40	89
Interface hepatitis + rosettes + emperipolesis + plasma cells	27	93
Rosettes + plasma cells	27	91
Rosettes + emperipolesis	44	88
Rosettes + interface hepatitis	44	82

Autoimmune Hepatitis – Review of Histological Features Included in Simplified Criteria

(Balitzer, Modern Pathology 2017)

88 cases of AIH (including acute AIH) versus 20 cases of PBC and 13 of non-autoimmune acute hepatitis

Table 3 Pathologic characteristics of autoimmune hepatitis, primary biliary cholangitis and non-autoimmune acute hepatitis

	Autoimmune hepatitis (n = 88)	*Primary biliary cholangitis (n = 20)*	*Non-autoimmune acute hepatitis (n = 13)*	P-*value (autoimmune hepatitis vs primary biliary cholangitis, autoimmune hepatitis vs non-autoimmune acute)*
Emperipolesis	57 (65)	10 (50)	10 (77)	0.31 0.53
Rosettes	29 (33)	0	5 (38)	0.78
Plasma cells (any number)	87 (99)	14 (70)	11 (85)	< 0.001 0.044
Numerous plasma cells	43 (49)	4 (20)	1 (8)	0.024 0.006
Plasma cell clusters	49 (56)	7 (35)	2 (15)	0.582 0.008

- Emperipolesis and rosettes lack diagnostic specificity for AIH
- Plasma cells (any or numerous) more frequent in AIH

Histologic Features of Autoimmune Hepatitis: A Critical Appraisal

(Gurung, Human Pathology 2018)

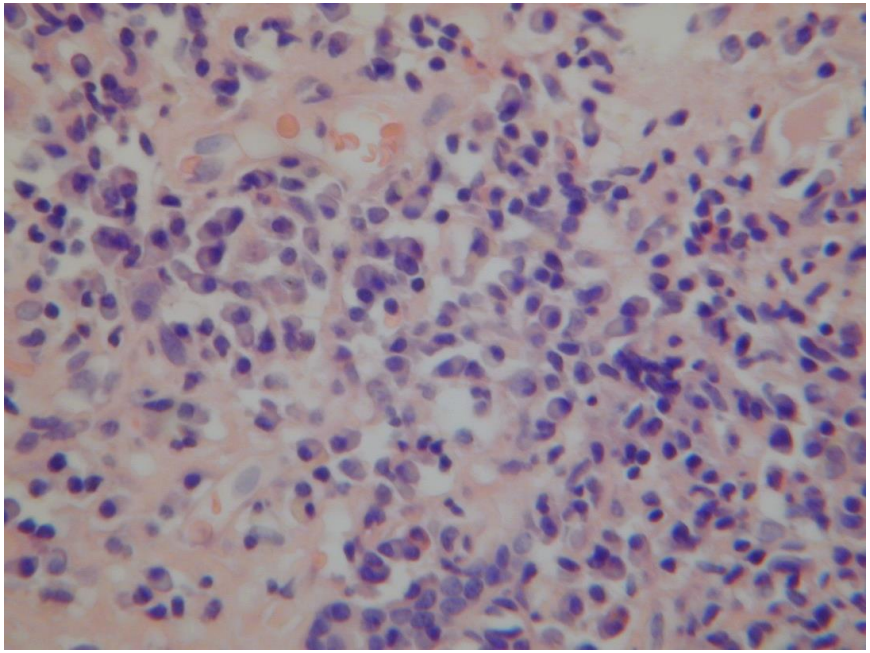
Liver biopsies from patients with untreated AIH (43) and hepatitis C (42)

Histological feature	HCV (n = 42)	AIH (n=43)	*P*
Rosettes *	7 (17%)	16 (37%)	.04997
Emperipolesis *	10 (24%)	22 (51%)	.01
Inflammatory grade - Metavir 0-2	25 (60%)	15 (35%)	.03
Inflammatory grade – Metavir 3-4	17 (40%)	28 (65%)	.03
Plasma cell clusters - portal	11 (26%)	26 (60%)	.002
Plasma cell clusters - lobular	2 (5%)	15 (35%)	.001

* Rosettes and Emperipolesis (AIH vs HCV)

- No significant difference when biopsies matched for inflammatory grade

Chronic Autoimmune Hepatitis -Typical Features
Plasma cell rich portal inflammation



BUT:

1. Plasma cells not essential to support/confirm diagnosis of AIH
   - Up to 1/3rd of cases have few/no plasma cells (EASL Guidelines 2015)

2. Plasma cells also seen in other diseases associated with portal inflammation:
   - Chronic biliary disease – e.g. PBC
     - mainly IgM+ PCs in PBC (vs IgG+ in AIH) (Cabibi 2010, Moreira 2010, Abe 2014)
   - Acute hepatitis – e.g. Hepatitis A

Autoimmune Hepatitis – Laboratory Investigations
Diagnostic Criteria

- **Diagnostic algorithms / scoring systems mainly designed for adults presenting with "typical" chronic AIH**

- **Problems with applying similar approaches to other situations**
  - Acute / fulminant AIH
  - Paediatric AIH
  - Drug-induced AIH
  - "Overlap syndromes" – e.g. PBC, PSC, IgG4 disease
  - Concurrent disease – e.g. NAFLD, HCV

Autoimmune Hepatitis - Acute Presentation
Incidence & Diagnostic Criteria

30- 40% of cases present as acute hepatitis /acute liver failure

(Manns 2010, Lohse 2011, Gleeson 2012, EASL Guidelines 2015)

Autoantibodies/immunoglobulins unreliable in the diagnosis of acute AIH

- Autoantibodies and hypergammaglobulinaemia may not be present at the time of presentation with acute AIH (Lohse 2011, Czaja 2013, EASL Guidelines 2015, Fujiwara 2016)
  - 30-40% have absent or weakly positive ANA
  - 25-40% have normal IgG

- Autoantibodies present in up to 40% of patients with other causes of acute liver failure - e.g viral or drug-induced (Bernal 2007)

Acute Presentation of Autoimmune Hepatitis - Histological Features

1. Acute presentation of chronic liver disease

- 14-35% have features of chronic hepatitis (Fujiwara 2011, Yasui 2011, Fujiwara 2016, Dohmen 2017)
- 2-95% have fibrosis /cirrhosis (Nikias 1994, Burgart 1995, Miyake 2010, Fujiwara 2011, Nguyen Canh 2017, Joshita 2018)

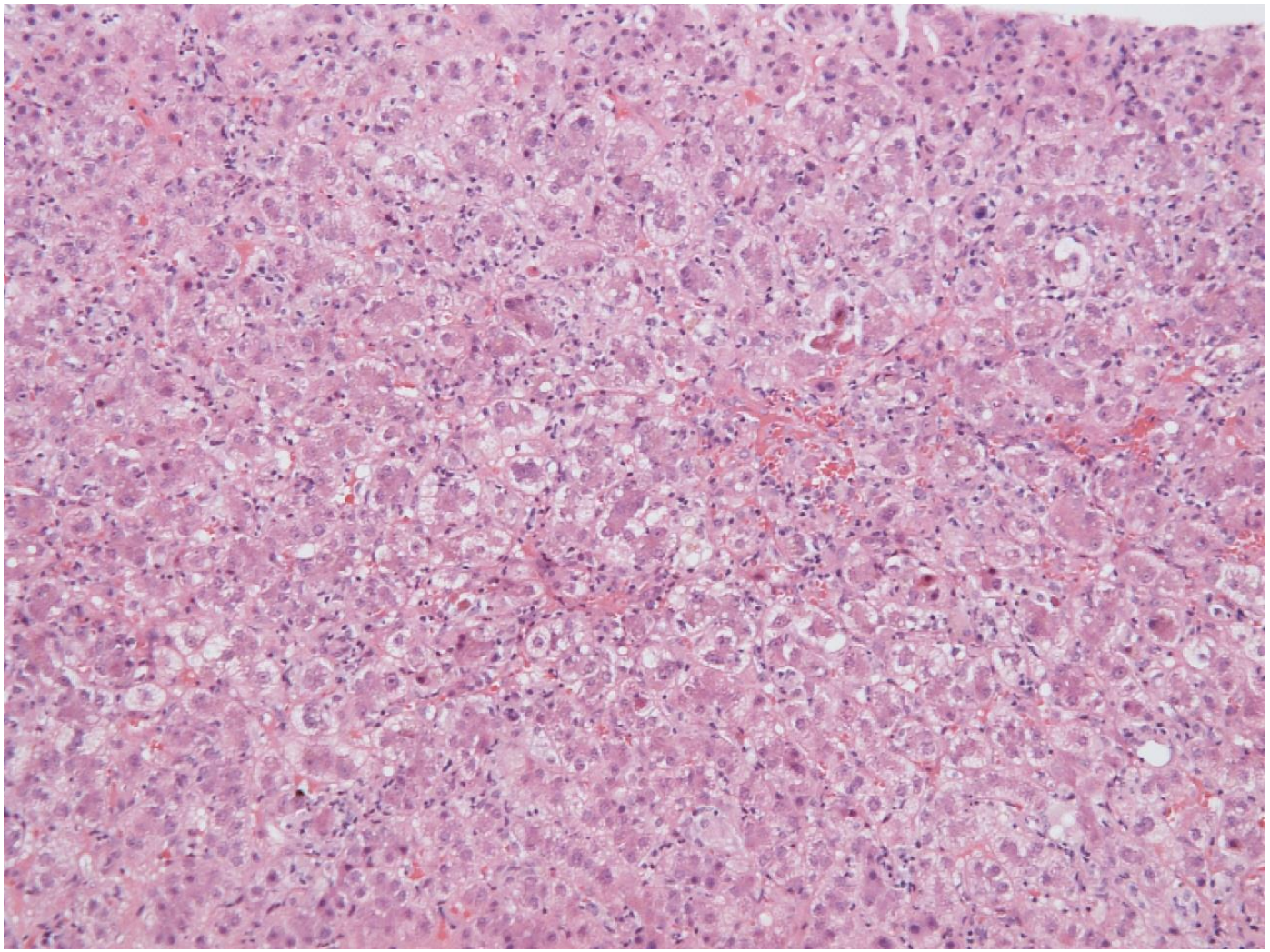
2. Acute hepatitis (with no signs of chronic liver disease)

- Classical features of acute lobular hepatitis (resembling viral or drugs)

Acute Autoimmune Hepatitis

Diffuse lobular inflammation

ballooning, lobular disarray and spotty necrosis



Acute Lobular Hepatitis - Histological Features Favouring a Diagnosis of AIH

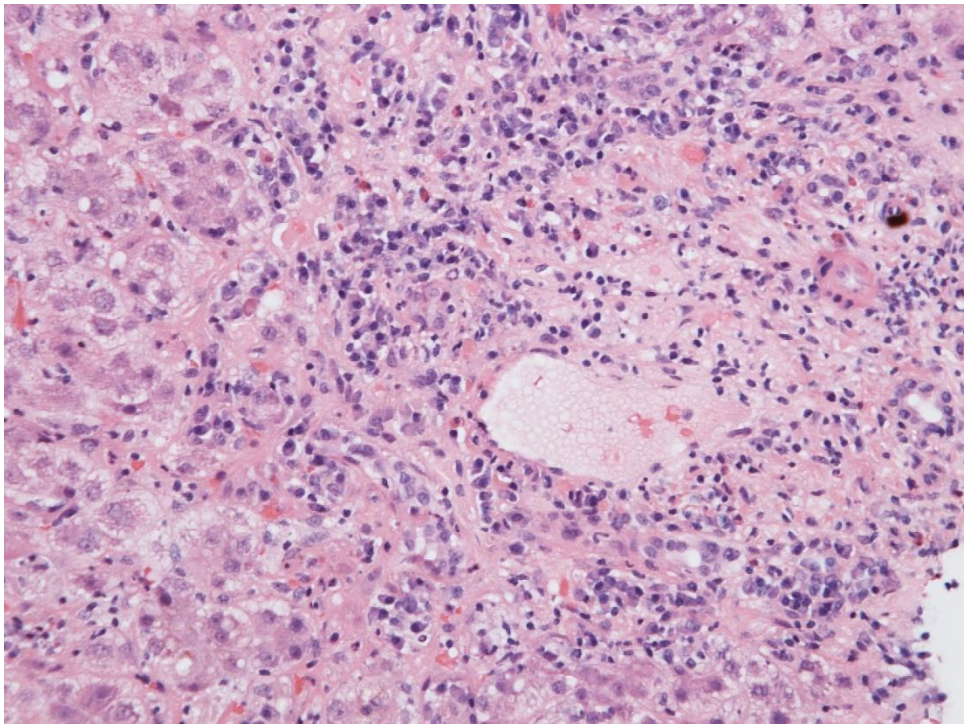
(Abe 2007, Fujiwara 2008, Stravitz 2011, Susuki 2011Yasui 2011, Fujiwara 2016, Dohmen 2017, Nguyen Canh 2017)

- Portal inflammation / interface hepatitis (resembling chronic AIH)
- Plasma-cell rich inflammatory infiltrate (portal and lobular)
- Portal lymphoid follicles
- Centrilobular inflammation & necrosis / central perivenulitis
- Hepatocyte rosettes
- Emperipolesis
- Fibrosis

BUT

- None of the above features can be regarded as specific for AIH

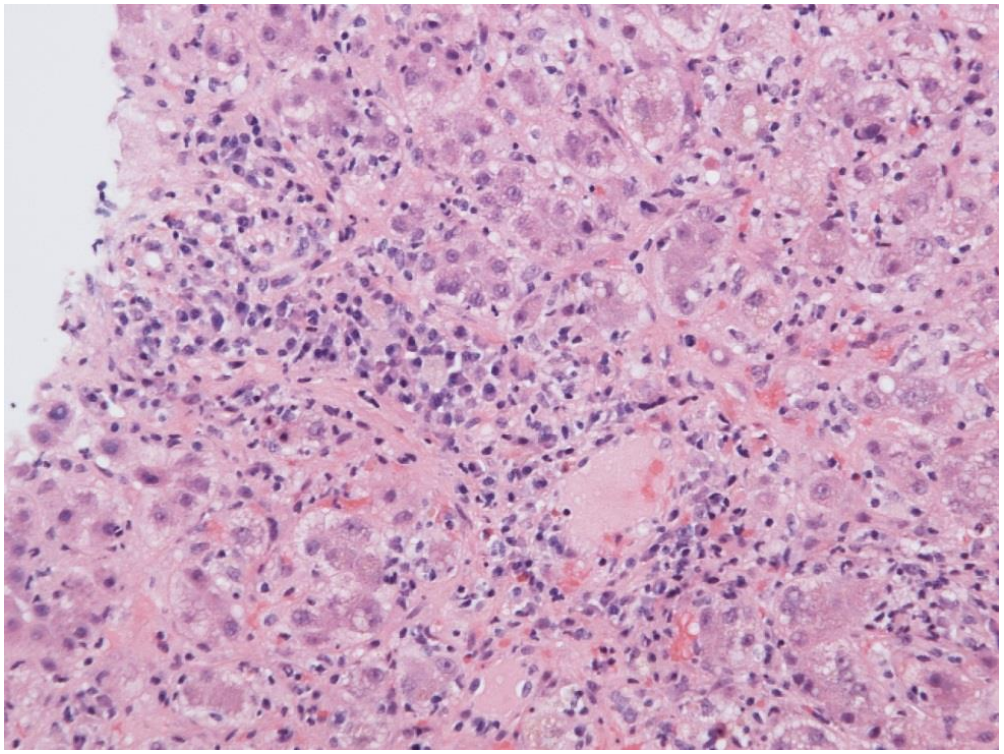
Portal Inflammation in Acute Autoimmune Hepatitis



1. **Portal inflammation occurs in all cases of acute hepatitis (viral, drug & AIH)**
   - Plasma cell rich infiltrate favours a diagnosis of AIH

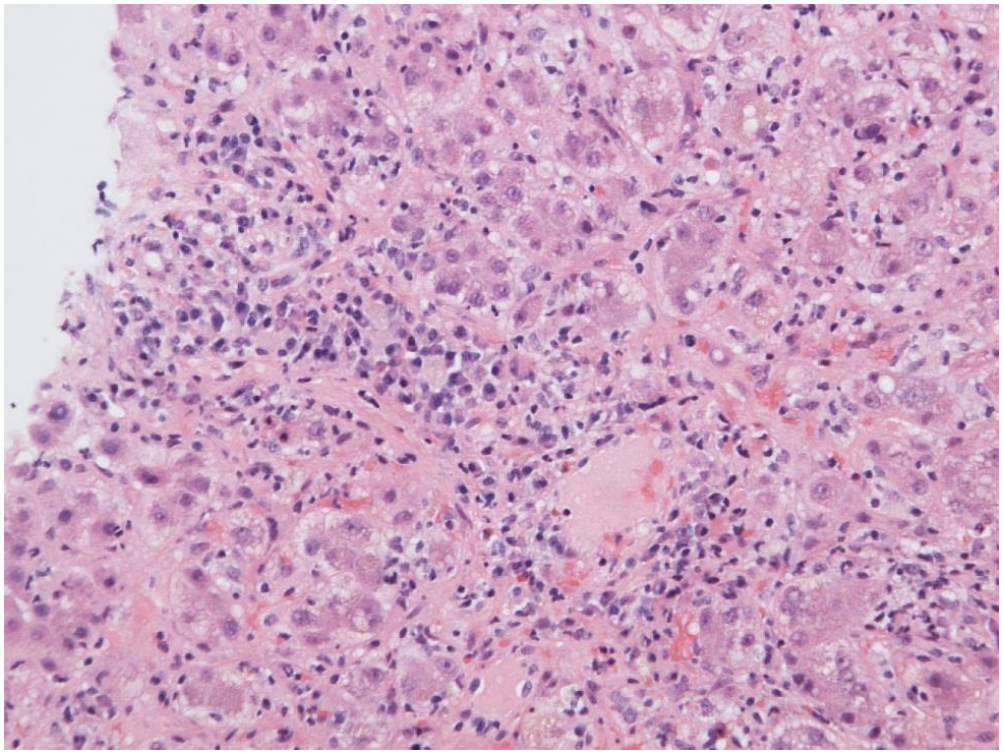
2. **Interface inflammation resembles interface hepatitis seen in chronic AIH**
   - But interface inflammation difficult to assess in the presence of diffuse lobular inflammation

Centrilobular Necroinflammatory Changes ("Central Perivenulitis") in AIH



- Usually occurs as centrilobular accentuation of diffuse lobular hepatitis - typically associated with portal inflammation
- Some cases may present as isolated central central perivenulitis – without diffuse lobular inflammation or portal inflammation
  - Lower frequency/titre of ANA, lower levels of IgG
  - Good response to treatment with immunosuppression (Aizawa 2016)

Centrilobular Necroinflammatory Changes ("Central Perivenulitis") in AIH



- Similar changes may occur in other forms of acute lobular hepatitis – e.g. viral, drugs
- Plasma cell rich infiltrates may be a pointer
- Hepatocyte rosetting and emperipolesis ("typical" features of chronic AIH) unhelpful in this setting
  - commonly seen in non-autoimmune acute hepatitis (Balitzer, Modern Pathology 2017)

Autoimmune Hepatitis Presenting as Acute Liver Failure

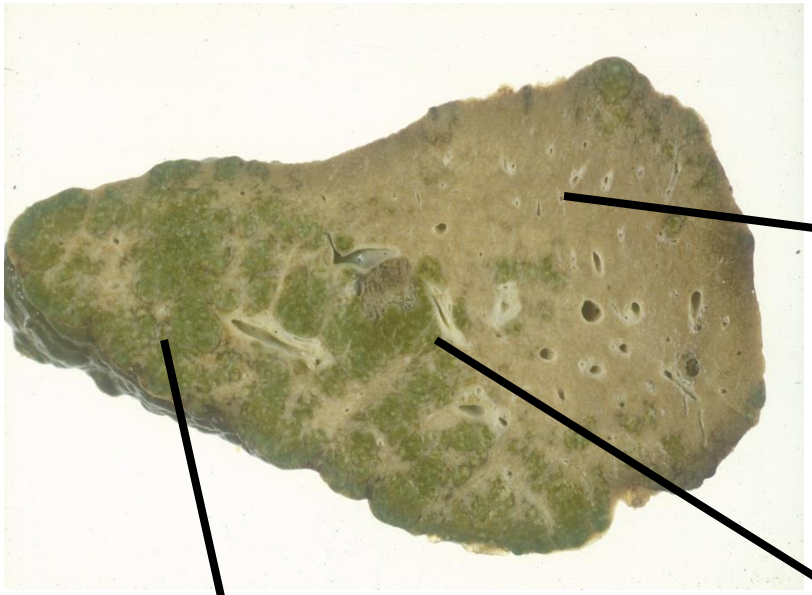
Histological Features

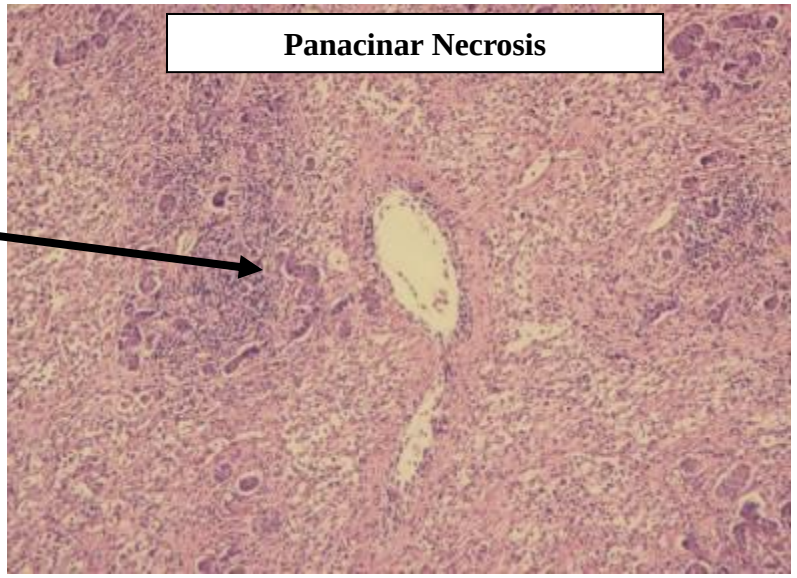
- Severe cases have panacinar / multiacinar necrosis
- Many patients are too sick to have a liver biopsy

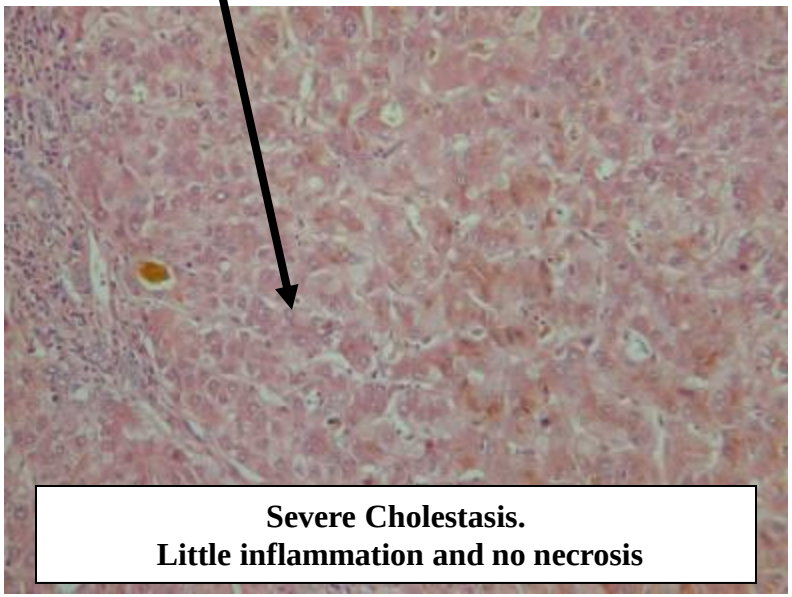
Problems with liver biopsy assessment

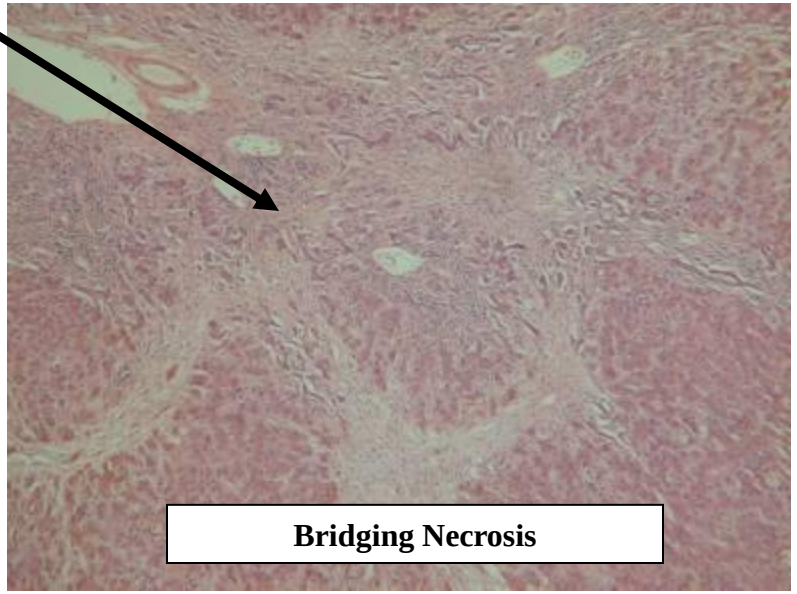
- Changes heterogeneous in distribution (sampling variability)
- Typical features of AIH may no longer be apparent
- Can resemble changes seen in cirrhosis (histologically and radiologically)

Liver Transplantation for Subacute Liver Failure (Autoimmune Hepatitis)
Severe Acute Hepatitis with Submassive Hepatic Necrosis

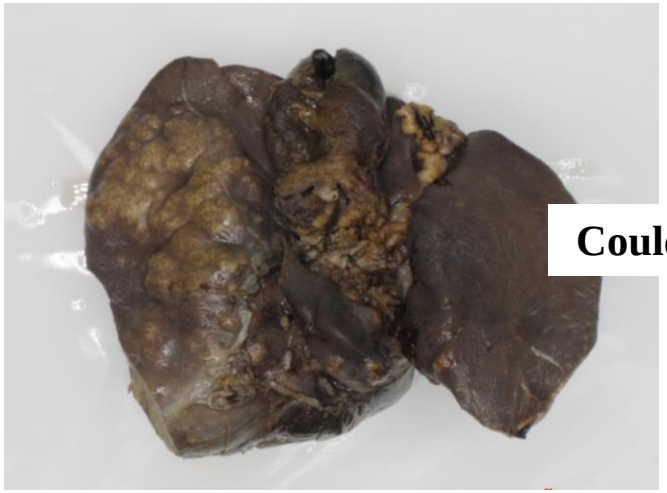






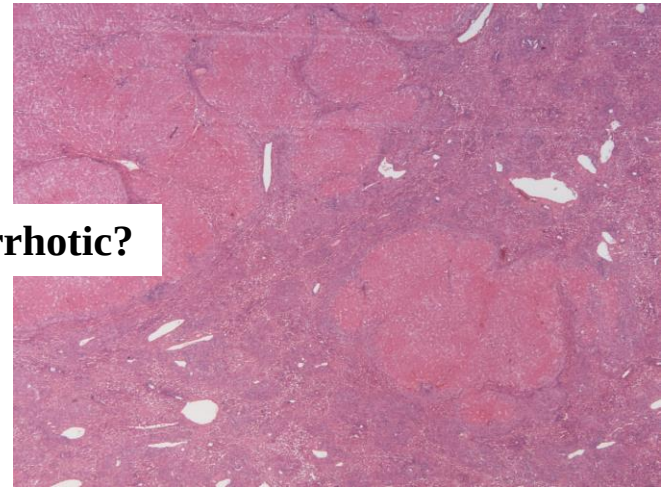


Liver Transplantation for Subacute Liver Failure (? Autoimmune Hepatitis)
Severe Acute Hepatitis with Submassive Hepatic Necrosis

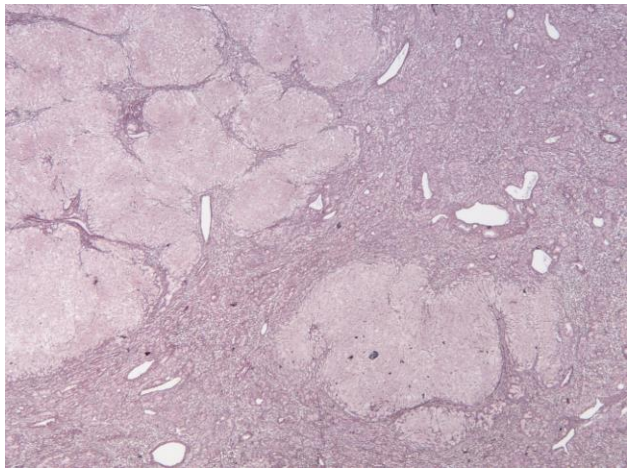


Shrunken nodular liver

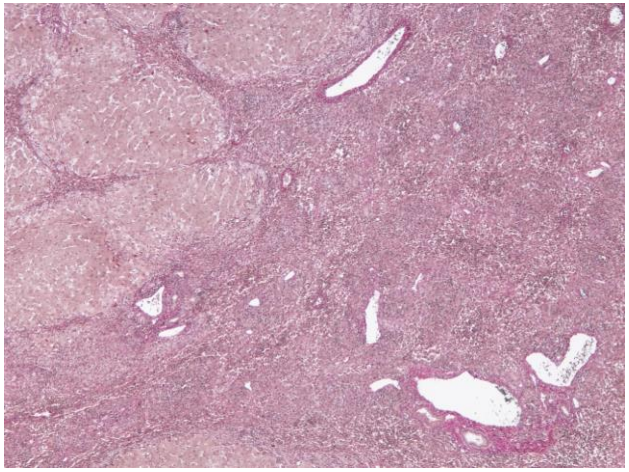
Could this be cirrhotic?



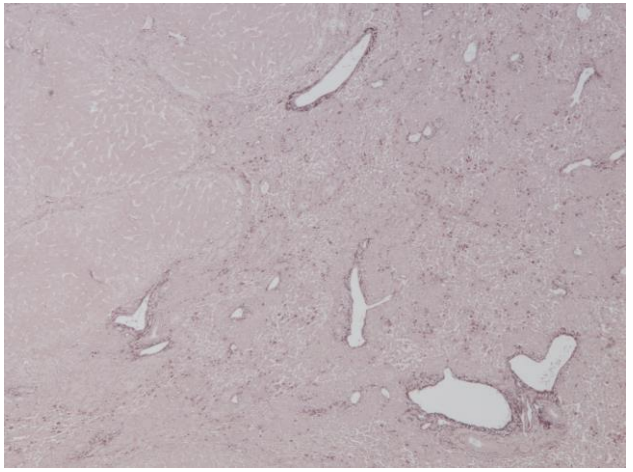
Hepatocyte nodules surrounded by inflamed fibrous tissue



Reticulin
Collapse in areas of bridging necrosis



HVG
Immature collagen fibres in areas of collapse



Orcein
No elastic fibres in areas of collapse

Role of Liver Biopsy in AIH

1. Establishing the diagnosis

2. **Assessing disease severity**
   - **Inflammatory activity**
     - **Interface hepatitis**
     - **Lobular inflammation**
   - Fibrosis

Chronic Autoimmune Hepatitis
Interface Hepatitis - Clinical Significance

Prognosis

- Presence/severity at presentation predicts development of fibrosis
- Persistence after treatment associated with increased risk of relapse (Manns 2010)
  - Interface hepatitis present in up to 50% of patients with normal transaminases and IgG (Luth 2008).

Treatment

Newly Diagnosed AIH

- Indication for commencing immunosuppression
- Mild activity (e.g. Ishak score <4-6) in older person may be grounds for not treating with immunosuppression (BSG Guidelines - Gleeson 2011)

Treated AIH (Absence of IFH = main criterion for defining "histological remission")

- Presence of ongoing IFH is indication for maintaining immunosuppression
- Histological response typically lags several months behind biochemical response
- Plasma cells and severe fibrosis may also be predictive of relapse in the absence of interface hepatitis (Czaja 2003, Czaja 2014)

Lobular Inflammation in Autoimmune Hepatitis
Clinical Significance

Acute Presentation

- Presence of extensive bridging necrosis or panacinar necrosis associated with increased risk of acute liver failure.
- Role of immunosuppressive therapy in this setting is uncertain.

Chronic AIH

- Presence of confluent / bridging necrosis associated with high risk of progression to cirrhosis - up to 80% (Cjaza 2007, Manns 2010)
- Strong/absolute indication for immunosuppression - but less responsive to immunosuppression than cases with pure interface hepatitis

Role of Liver Biopsy in AIH

1. Establishing the diagnosis

2. **Assessing disease severity**

   - Inflammatory activity

   - **Fibrosis**

     - **Non-invasive markers (e.g. Fibroscan) less well established in AIH compared with other chronic liver diseases (e.g. HCV, NAFLD)**

     - **Liver stiffness also affected by inflammation**

Autoimmune Hepatitis – Assessment of Fibrosis

25-33% of adults and >50% of children have cirrhosis at presentation
(Lohse 2011, Gleeson 2012, Floreani 2013, Kirsten 2015, EASL Guidelines 2015)

- Includes cases with acute presentation

Patients with cirrhosis at presentation

- Have worse outcome (Feld 2005, Verma 2007, Landeira 2012, Kirsten 2015)
- Less responsive to immunosuppression (Muratori 2009, Efe 2012, Anand 2018)
  - Some studies suggest that cirrhosis at diagnosis does not predict treatment responses or adverse outcomes (Yeoman 2011, Yoshisawa 2012,Ngu 2013)
  - Reversal of fibrosis and cirrhosis can occur following treatment (Czaja 2014, Borssen 2016)

- At risk of developing HCC - approx 0.1 -1%/year (Yeoman 2008, Migita 2012, Hino-Arinaga 2012, Gronbaek 2014, Tansel 2017)
  - Screening as for other patients with cirrhosis
  - Occasional cases of HCC occur in non-cirrhotic AIH (Valean 2019)

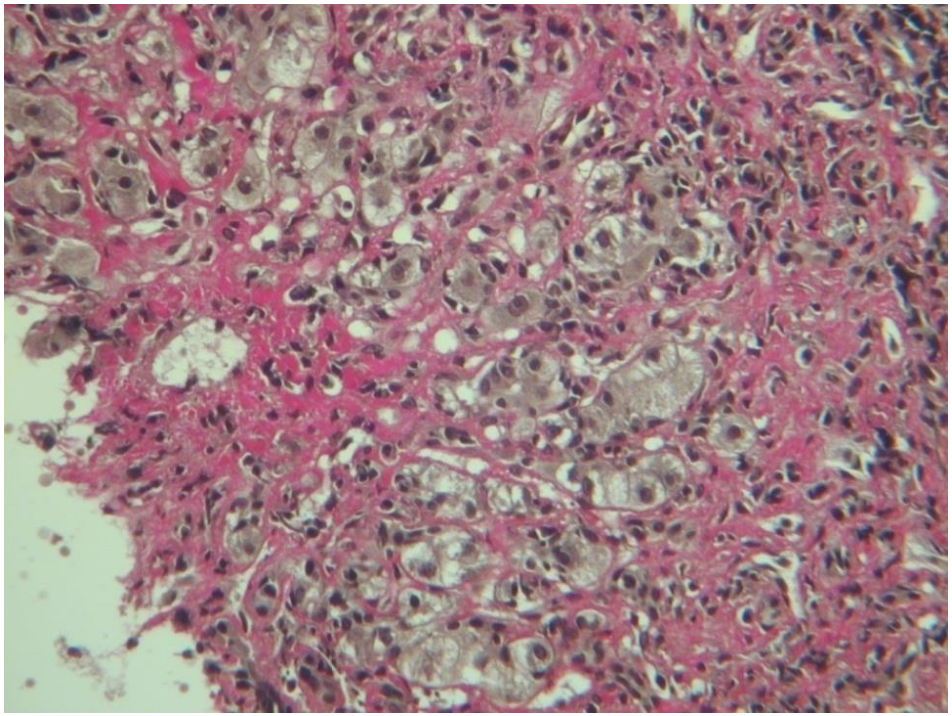
Role Of Liver Biopsy in AIH
Problems with Assessing Fibrosis/Cirrhosis

1. Areas of bridging/panacinar necrosis and nodule formation in severe acute hepatitis can produce changes resembling those seen in cirrhosis.
   - Use of connective tissue stains to distinguish recent collapse from longstanding fibrosis.

2. Cirrhosis in AIH often irregular and/or macronodular
   - Sampling problem with liver biopsy
   - Possible role of laparoscopy to identify macronodules and obtain suitable tissue samples (Denzer 2007, Lohse & Mieli-Vergani 2011, EASL Guidelines 2015)

3. Distinctive pattern of lobular "dissecting fibrosis" – difficult to classify or quantify according to current histological staging systems.

Autoimmune Hepatitis - Lobular Dissection by Fibrous Tissue



- Delicate strands of fibrous tissue extending from portal tracts to hepatic veins
- Surround and separate small clusters of hepatocytes forming rosettes
- Normal vascular relationships retained, no elastic fibres
- Distinctive pattern – different to post-necrotic collapse and cirrhotic septa

Mechanism uncertain

- May be a consequence of severe interface hepatitis and/or lobular inflammation
- Possibly reflects acute evolving to chronic liver injury

Role of Liver Biopsy in the Diagnosis and Management of AIH
Summary & Conclusions

1. Liver biopsy continues to play an important role in the diagnosis and management of AIH.

2. Some features regarded as typical for AIH (rosettes, emperipolesis) may have problems with reproducibility and diagnostic specificity.

3. Diagnostic criteria and scoring systems devised for “typical cases” of chronic AIH may not be applicable to other situations (e.g. acute AIH, paediatric AIH, drug-induced AIH, “overlap syndromes”, patients with concurrent diseases)

4. Assessment of the severity of inflammatory activity and fibrosis has implications for prognosis and treatment.