BSG/ACP annual course
Royal College of Pathologists

Infections and Inflammations of the Hepato-Biliary System

Hepatitis B and D

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Thurs 10 Dec 2009

Hepatitis B and D
Outline

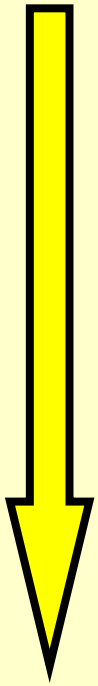
- HBV
  - Acute HBV
  - Chronic HBV
  - Role of biopsy in management of chronic HBV infection
    - Grading & staging
      - Adequacy of biopsy
    - HBV special stains and immunohistochemistry
    - Additional and alternative conditions encountered in patients with chronic HBV hepatitis
- HDV
- Duties of the pathologist regarding an HBV/HDV liver biopsy

HBV infection

- **42 nm incompletely double stranded DNA Hepadnavirus with HBsAg envelope**
- **Leading cause worldwide of chronic liver disease and HCC**
- **350x10^6 chronically infected**
- **Up to 15% of population infected in China, SE Asia & sub-Saharan Africa**
- **Transmission: vertical, horizontal (children), sexual, parenteral**

HBV infection

Acute HBV infection



65% asymptomatic

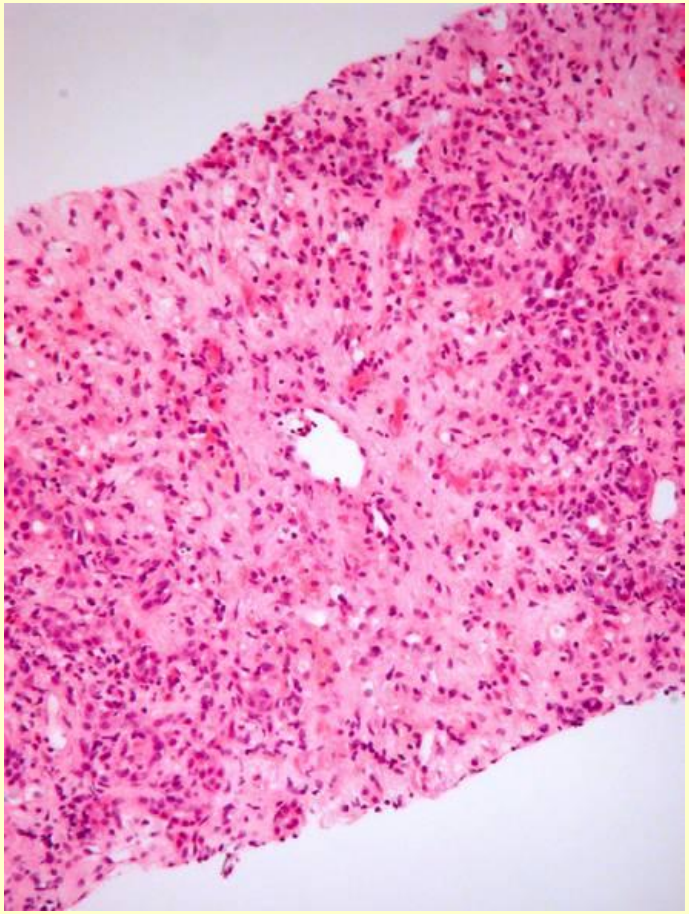
35% Flu-like illness Nausea Vomiting Jaundice

<1% fulminant liver failure

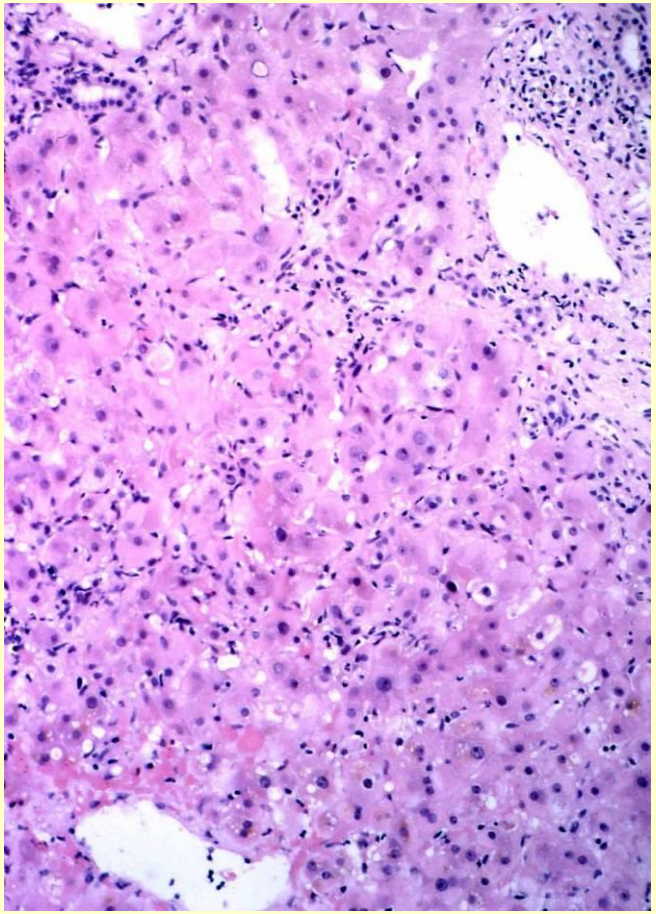
Acute fulminant hepatitis

≤5% acquire chronic infection
of whom ~30% develop cirrhosis
of whom ~10% develop HCC

Acute HBV infection



Acute fulminant hepatitis
(massive hepatic necrosis)



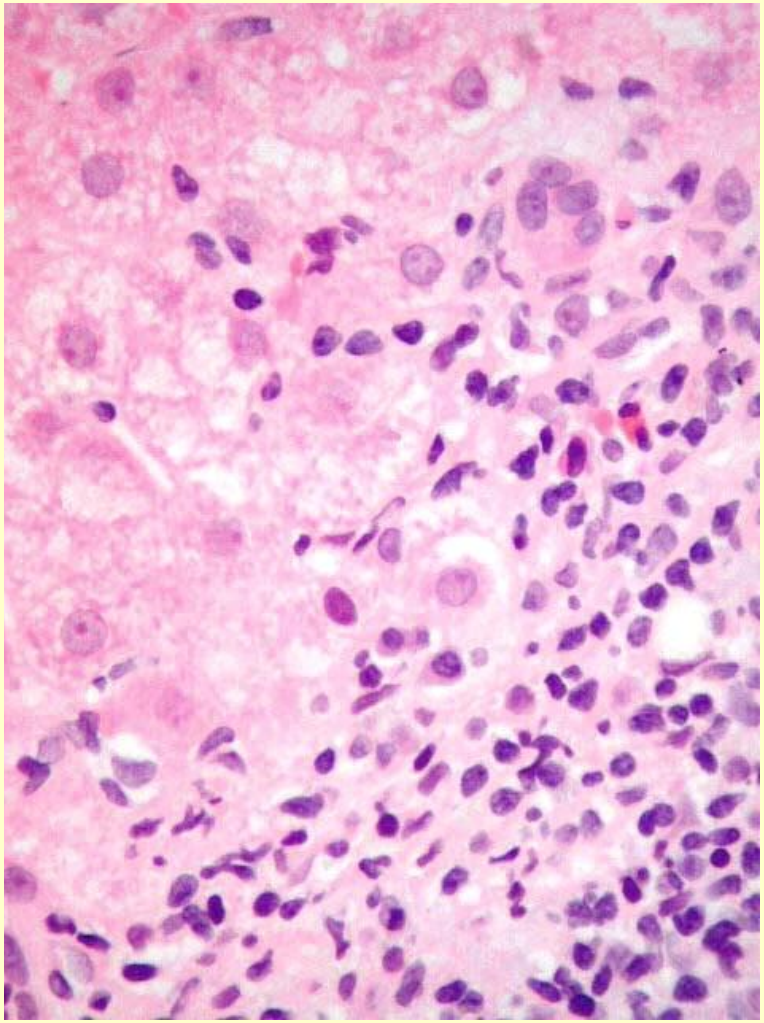
Acute recurrent HBV
post transplantation

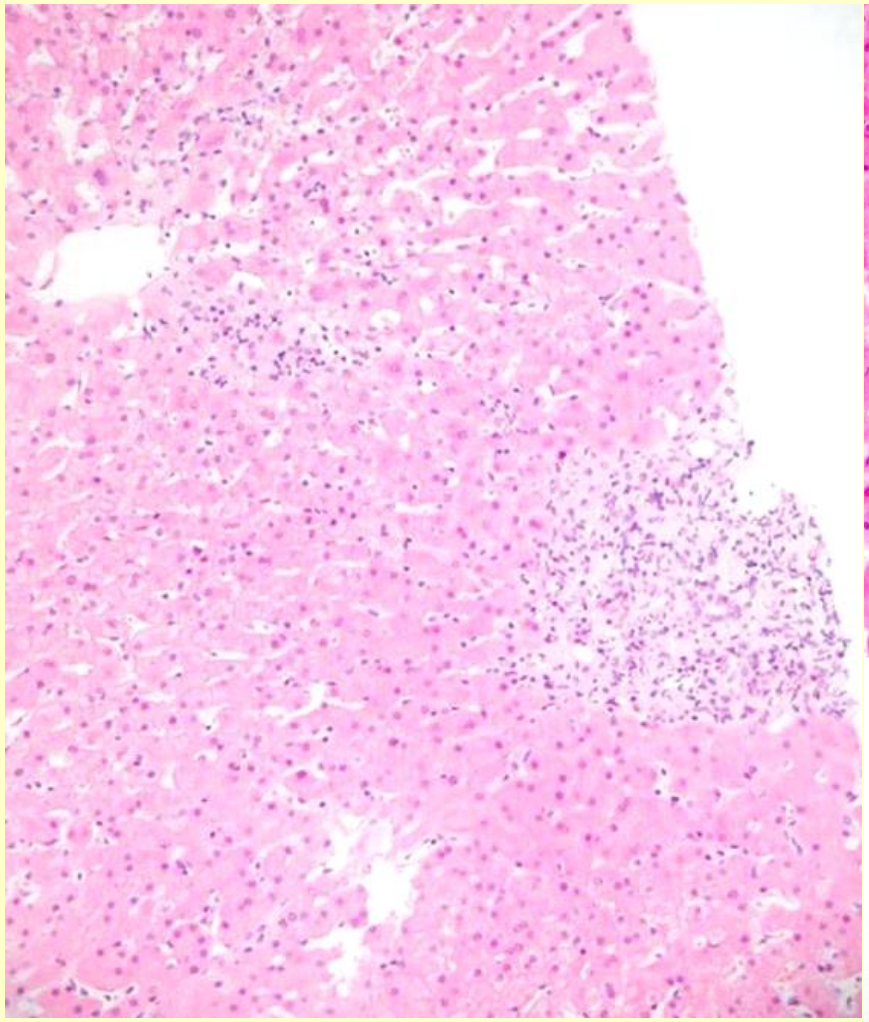
Reasons to perform liver biopsy during chronic HBV infection.

- To confirm that the suspected liver disease is due to chronic HBV hepatitis and to assess HBV disease activity, and to define alternative and additional diagnostic possibilities
- The decision to institute treatment is based on observations such as:
  - "Factors that predict responsiveness to IFN include high baseline histologic activity index and low HBV DNA."
  - "Pretreatment factors that predict the response to lamivudine are very similar to those for IFN except that baseline HBV DNA may not be as important a predictor for lamivudine efficacy."
    - Heathcote J. Semin Liv Dis 23, 69-79; 2003
- ie treatment is considered relatively ineffective in immunotolerant patients; but this may not be so with the newer nucleos/tide antivirals

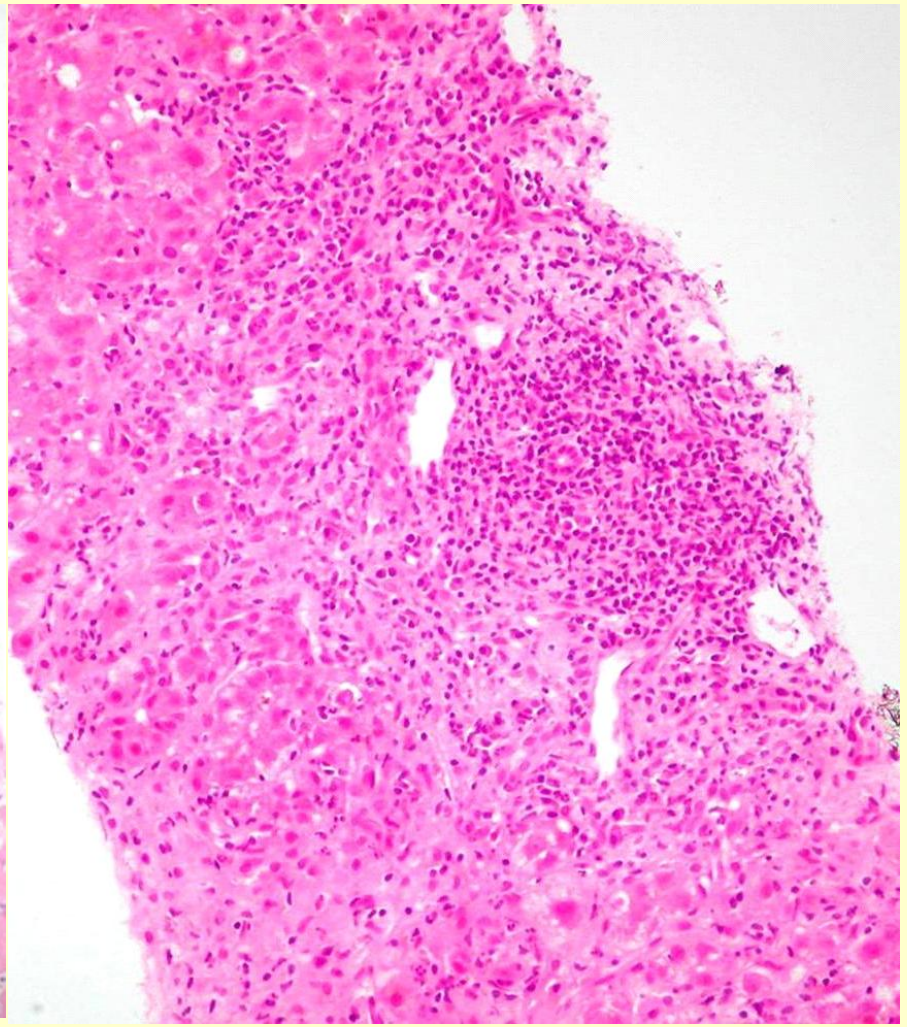
Piecemeal necrosis

- **“is characterised by lymphocytic infiltration and destruction of hepatocytes at the connective tissue-parenchymal interface around portal tracts and along the fibrous septa.”**
  - **Desmet V. Hepato-gastroenterol 38, 14-21; 1991**

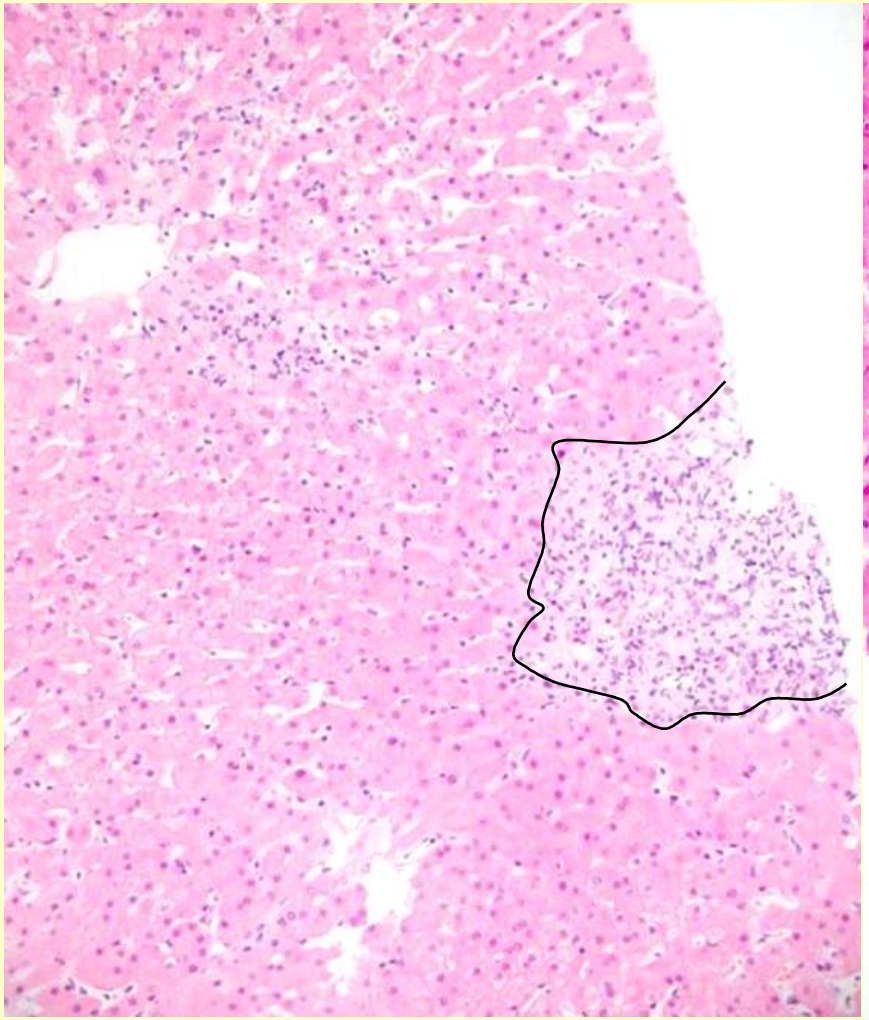




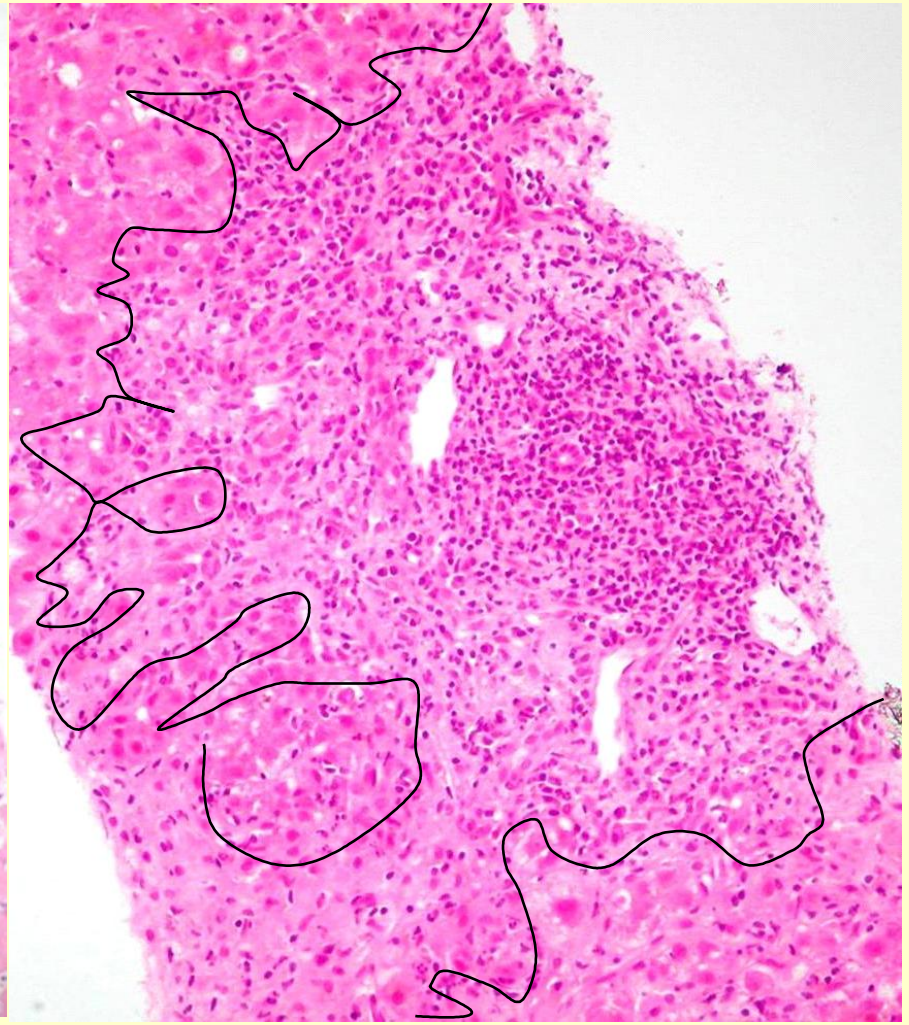
Mild piecemeal necrosis



Severe piecemeal necrosis



Mild piecemeal necrosis



Severe piecemeal necrosis

Assessment of activity of chronic HBV infection

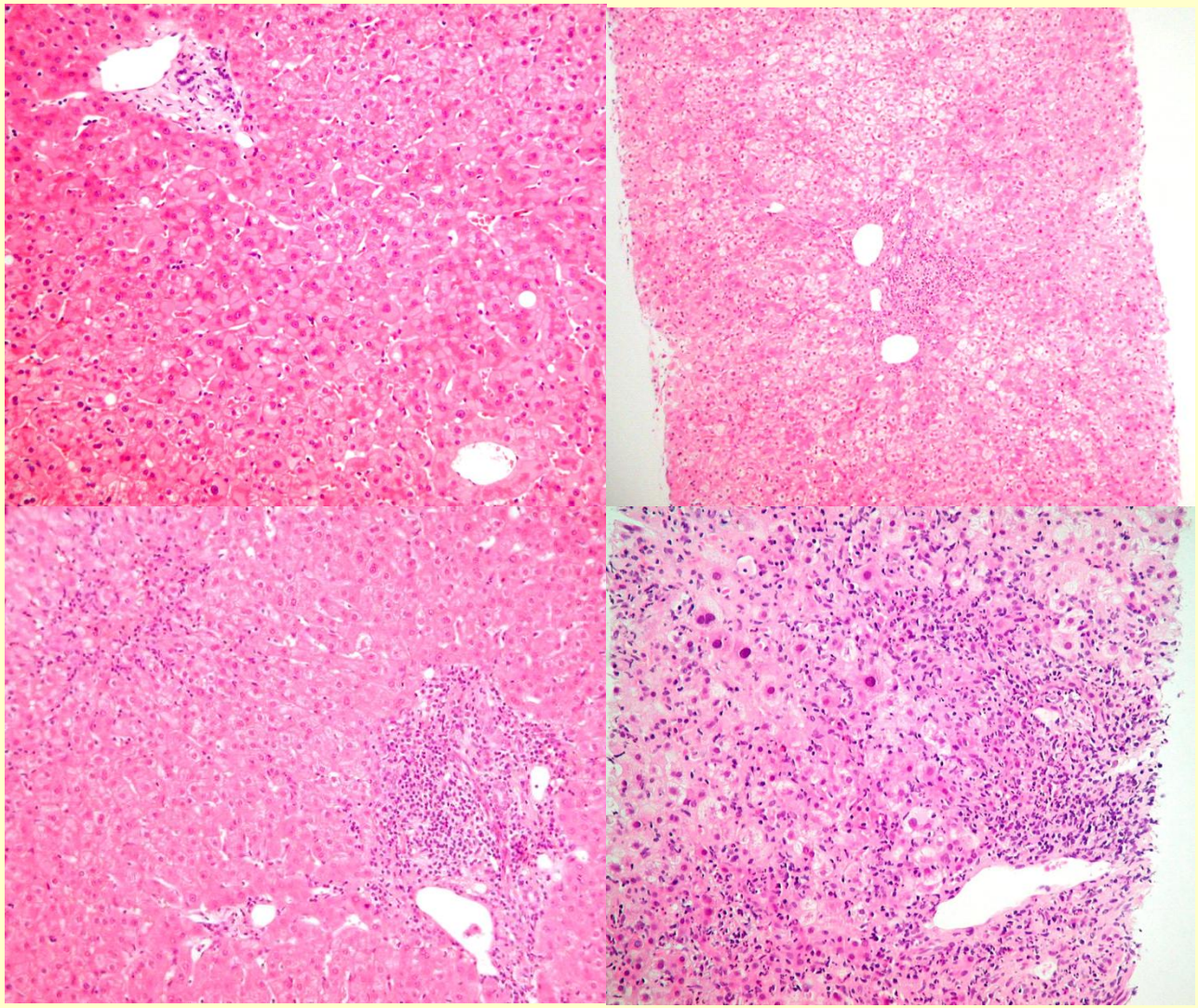
- **Currently, “activity” of chronic HBV hepatitis does not depend only on histopathology**
  - **Clinical disease “activity” depends on transaminase levels, viral load and viral replication, (as well as severity of inflammation and fibrosis in the liver biopsy)**
- **Histopathologically, chronic hepatitis activity does not depend solely on (piecemeal) interface hepatitis**
  - **Currently histopathological “necroinflammatory” hepatitis activity includes lobular and portal inflammation as well**
  - **Fibrosis and architectural changes also contribute to the overall “histological activity index” in chronic viral hepatitis**

Management of chronic viral hepatitis: grading and staging of hepatitis

- **Grade = inflammation**
  - **Lobular**
  - **Piecemeal**
  - **Portal**
- **Stage = fibrosis and architectural distortion**
  - **Fibrous expansion of portal tracts**
  - **Portal-portal links**
  - **Central-portal bridges**
  - **Nodules**

“Minimal” disease carrier

Mild

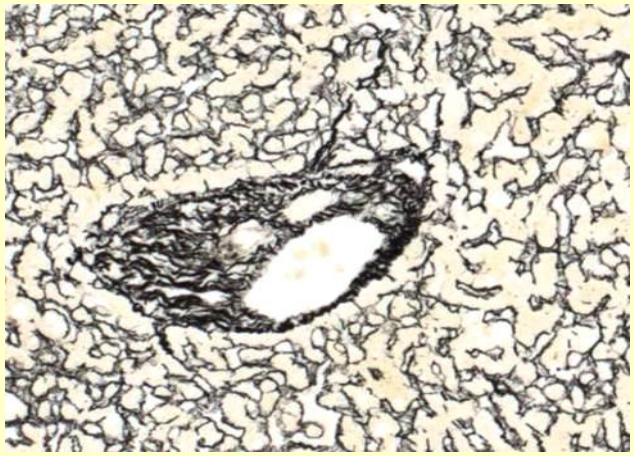


Moderate

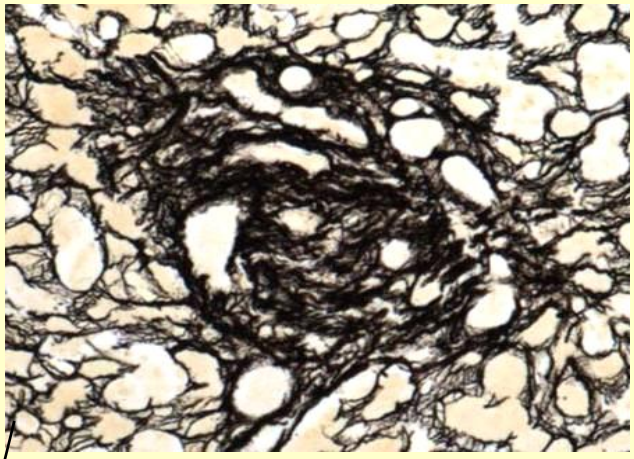
Severe

(Necro-inflammatory) grades of chronic HBV infection

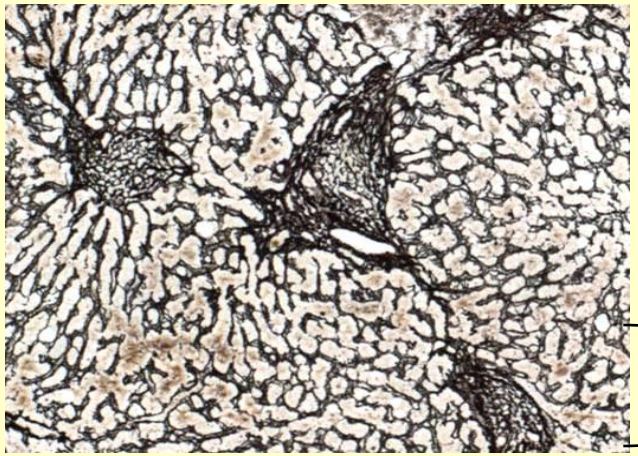
Staging of chronic hepatitis



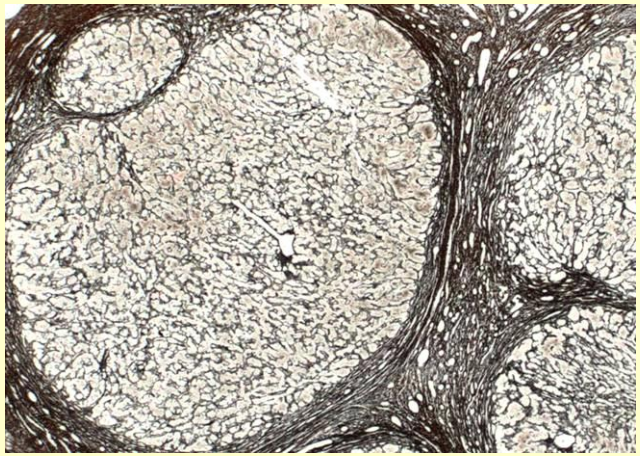
Normal



Expanded portal tract



Links between expanded portal tracts



Cirrhosis

Ishak scoring system

("modified histological activity index: Knodell updated")

J Hepatol 1995,22;696

Grading - A: Interface hepatitis (piecemeal necrosis)

0 Absent
1 Mild (focal, few portal areas)
2 Mild/moderate (focal, most portal areas)
3 Moderate (continuous, <50% tracts)
4 Severe (continuous, >50% tracts)

Grading - B: Confluent necrosis

0 Absent
1 Focal confluent necrosis
2 Zone 3 necrosis in some areas
3 Zone 3 necrosis in most areas
4 Zone 3 necrosis + occasional P-C bridging
5 Zone 3 necrosis + multiple P-C bridging
6 Panacinar or multiacinar necrosis

Grading - C: focal lytic necrosis, apoptosis, focal inflammation

0 Absent
1 One focus per x10 field
2 Two to four foci
3 Five to ten foci
4 More than ten foci

Grading - D: Portal inflammation

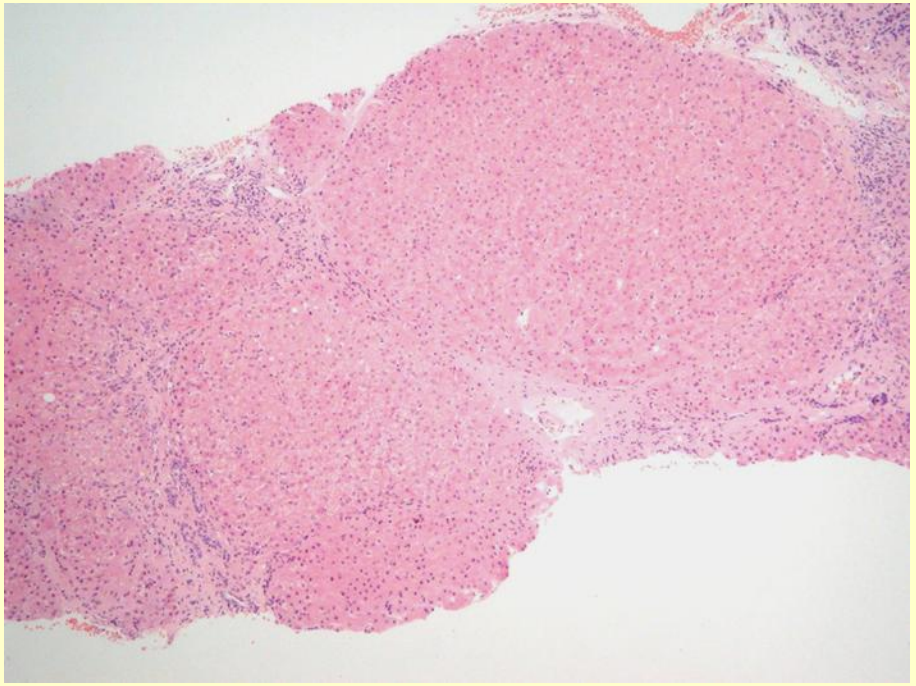
0 None
1 Mild, some or all portal areas
2 Moderate, some or all portal areas
3 Moderate/marked, all portal areas
4 Marked, all portal areas

Staging

0 No fibrosis
1 Some tracts expanded, +/- short septa
2 Most tracts expanded, +/- short septa
3 Most tracts expanded, some P-P bridges
4 Marked bridging, P-P and P-C
5 Marked bridging, occasional nodules (incomplete cirrhosis)
6 Cirrhosis, probable or definite

Aspects of the use of liver biopsy in chronic HBV infection.

- **Routine diagnosis requires descriptive reports, not scoring (major problems of intra-observer and inter-observer scoring variations in sporadic observations)**
  - **Standish R et al.**
    - **An appraisal of the histopathological assessment of liver fibrosis. Gut 55,569;2006**
- **The use of biopsy "scores" as numbers in clinical trials is wrong (and always has been)**



Which is the better description:
"Ishak grade 3 stage 6" or
"cirrhosis with mild inflammatory activity"?

Adequacy of liver biopsy in chronic hepatitis

- Traditional acceptance of biopsies with 6 portal tracts or biopsies ~1-1.5 cm long as adequate is incorrect
- (Most studies of biopsy adequacy have been done in HCV infected biopsies)
- Inadequate biopsies underestimate stage and grade of disease
  - Sampling error is an important concern.
- In most diffuse liver diseases examination of 11-15 complete portal tracts is necessary
  - ~20mm of a 1.4mm diameter biopsy
  - Progressively longer samples of thinner biopsies are needed
    - Guido M and Rugge M. Liver biopsy sampling in chronic viral hepatitis. Semin Liv Dis 24,89;2004

What is “significant” hepatic inflammation and fibrosis in the liver biopsy?

Table 1. Standardisation of terminology in chronic hepatitis B virus (HBV) infection

(Based on Lok AS et al. Gastroenterology 120, 1828; 2001).

Terms	Diagnostic criteria
Chronic hepatitis B	1 Serum HBsAg positivity longer than 6 months 2 Persistent or intermittent elevation of ALT/AST levels 3 Serum HBV DNA $>10^5$ copies/ml 4 Liver biopsy with a necroinflammatory score (**≥ 4**
Inactive HBsAg carrier	1 Serum HBsAg positivity longer than 6 months 2 HBeAg(-); anti-HBe(+) 3 Serum HBV DNA $<10^5$ copies/ml 4 Persistently normal serum ALT/AST levels 5 Liver biopsy* showing absence of significant inflammation (necroinflammatory score <4)
Resolved hepatitis B	1 Serum HBsAg(-); anti-HBc(+) 2 Normal serum ALT levels 3 History of known acute or chronic hepatitis B 4 Undetectable serum HBV DNA (hybridisation assays)**

* In these circumstances liver biopsy is optional.

** HBV DNA may be detectable using sensitive polymerase chain reaction assays.

ALT = alanine aminotransferase; anti-HBe = antibody to hepatitis B e antigen (HBeAg); anti-HBc = antibody to hepatitis B core antigen; AST = aspartate aminotransferase; HBsAg = hepatitis B surface antigen.

Assessment and management of chronic hepatitis B
Nikolai V Naoumov Clin Med JRCPL 2, 306–10; 2002.

What is “significant” hepatic inflammation and fibrosis in the liver biopsy?

Lok AS, Heathcote EJ, Hoofnagle JH.

Gastroenterology 120, 1828; 2001.

- **Management of hepatitis B: 2000 – summary of a workshop (NIDDK-AGA):**
  - **No scoring system is specified.**
  - **No cut-off between histologically mild hepatitis vs moderate-severe disease activity (in terms of either grade or stage scores) is specified.**
- **“At issue is what criteria should be used to define moderate-to severe disease.”**

What is "significant" hepatic inflammation and fibrosis in the liver biopsy?

Subsequent treatment guidelines are equally vague

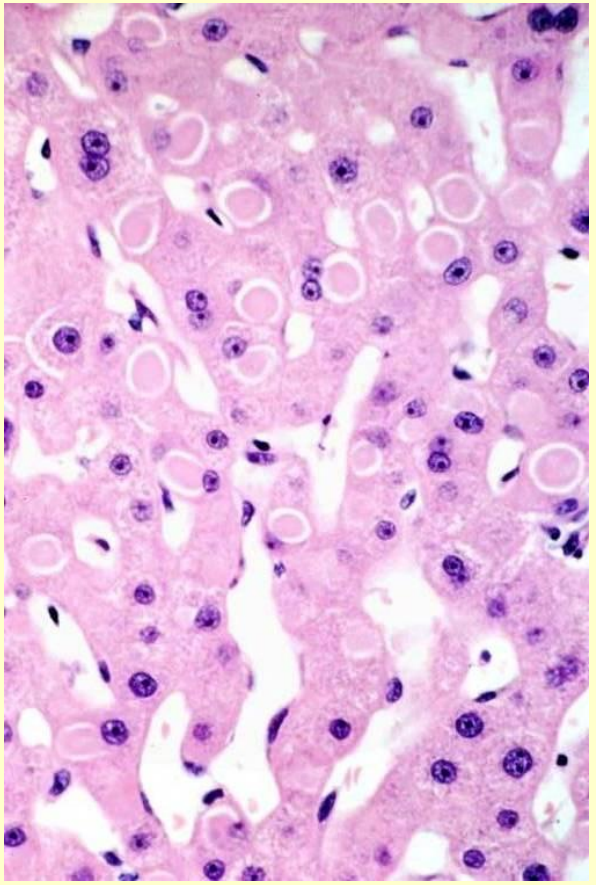
- Lok AS, McMahon BJ.
  - Chronic hepatitis B. Hepatology 34, 1225; 2001.
- EASL international consensus.
  - Conference on hepatitis B. J Hepatol 38, 533–540; 2003.
- Hadziyannis SJ et al.
  - Treatment of HBeAg–Negative Chronic Hepatitis B. Semin Liv Dis 23, 81-88; 2003.
- Heathcote J.
  - Treatment of HBe Antigen–Positive Chronic Hepatitis B. Semin Liv Dis 23, 69-79; 2003.
- Conjeevaram HS, Lok AS.
  - Management of chronic hepatitis B. J Hepatol 38, S90–S103; 2003.
- Lok AS, McMahon BJ.
  - Chronic hepatitis B: update of recommendations. Hepatology 39, 857; 2004.
- Keeffe EB et al.
  - A treatment algorithm for the management of chronic hepatitis B virus infection. Clin Gastroenterol Hepatol 2, 87; 2004.
- Liaw Y-F et al.
  - Asian-Pacific consensus statement on the management of chronic hepatitis B: a 2005 update. Liv Int. 25, 472; 2005.
- Lok A , McMahon B
  - AASLD practice guidelines. Hepatology 45,507;2007.
- Thomas HC.
  - Best practice in the treatment of chronic hepatitis B. J Hepatol. 47,588;2007.

EASL clinical practice guidelines: Indication for treatment
J Hepatol 50,227;2009

- The indication for treatment is mainly based on the combination of three criteria:
  - Serum HBV DNA levels
  - Serum ALT levels
  - Histological grade and stage
- Patients should be considered for treatment when:
  - HBV DNA levels are above 2,000 IU/mL
  - And/or the serum ALT levels are above the ULN
  - And/or liver biopsy shows moderate to severe active necroinflammation and/or fibrosis

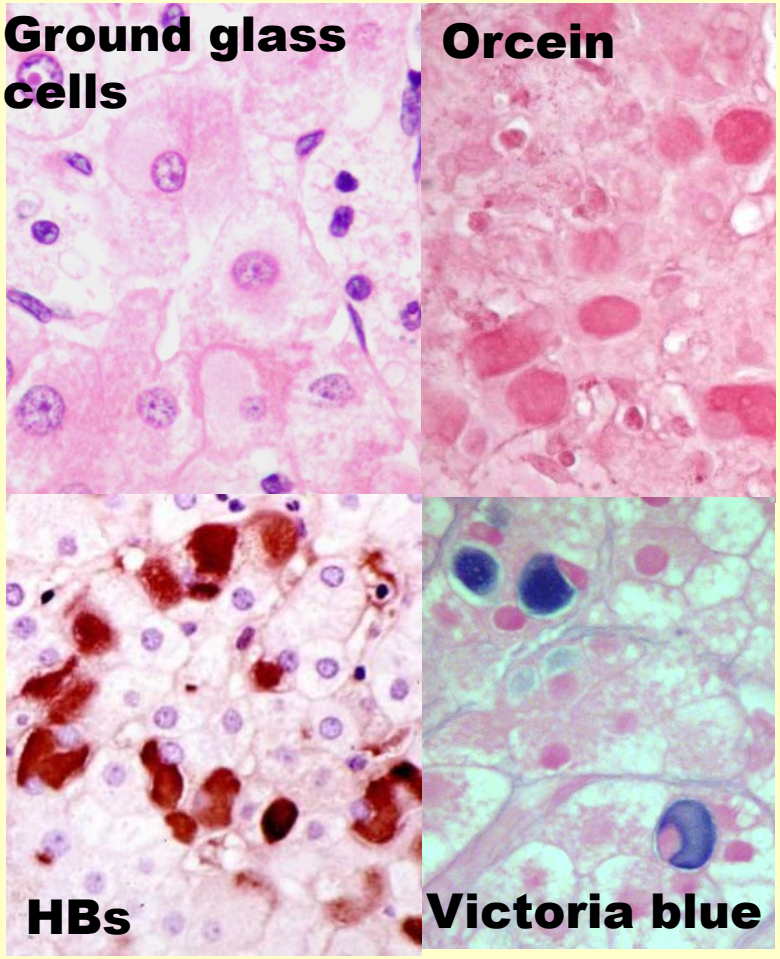
HBV special stains and immunohistochemistry

- **“Ground glass” hepatocytes contain endoplasmic reticulum full of HBsAg**
- **This cytoplasmic appearance can be simulated by:**
  - **Hepatocyte “oncocytosis”**
  - **Drugs: usually cyanamide toxicity is quoted but also various immunosuppressive agents and antibiotics may be implicated**
  - **Type IV glycogenosis**
  - **Lafora’s disease**
  - **Fibrinogen storage disease**
- **Therefore it’s best to assert that there are ground glass hepatocytes in the context of HBV infection after you have seen the confirmatory HBV stains**

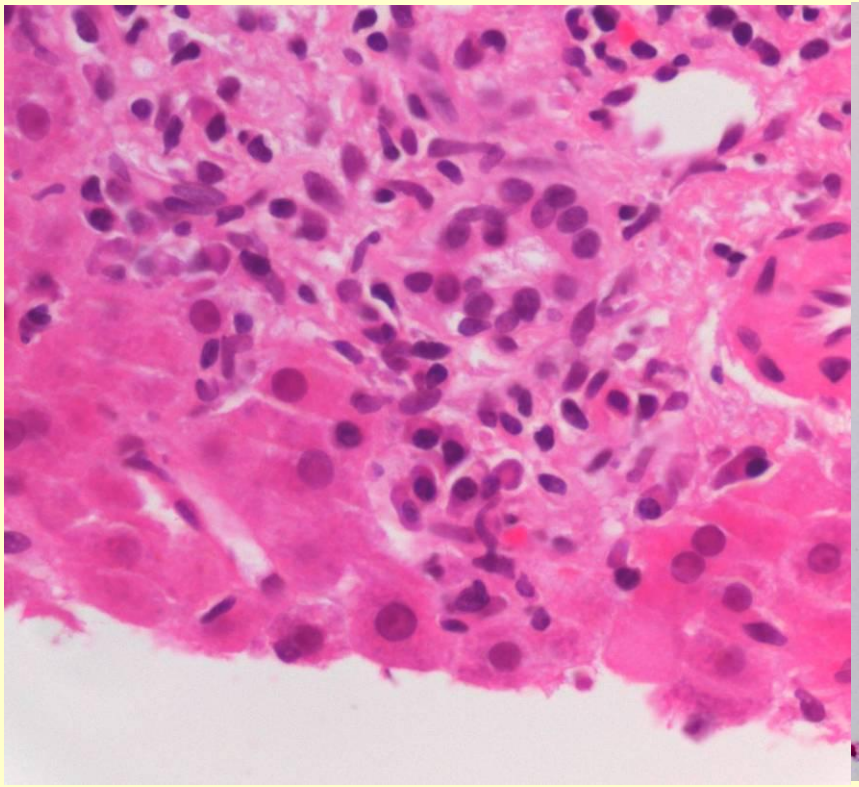


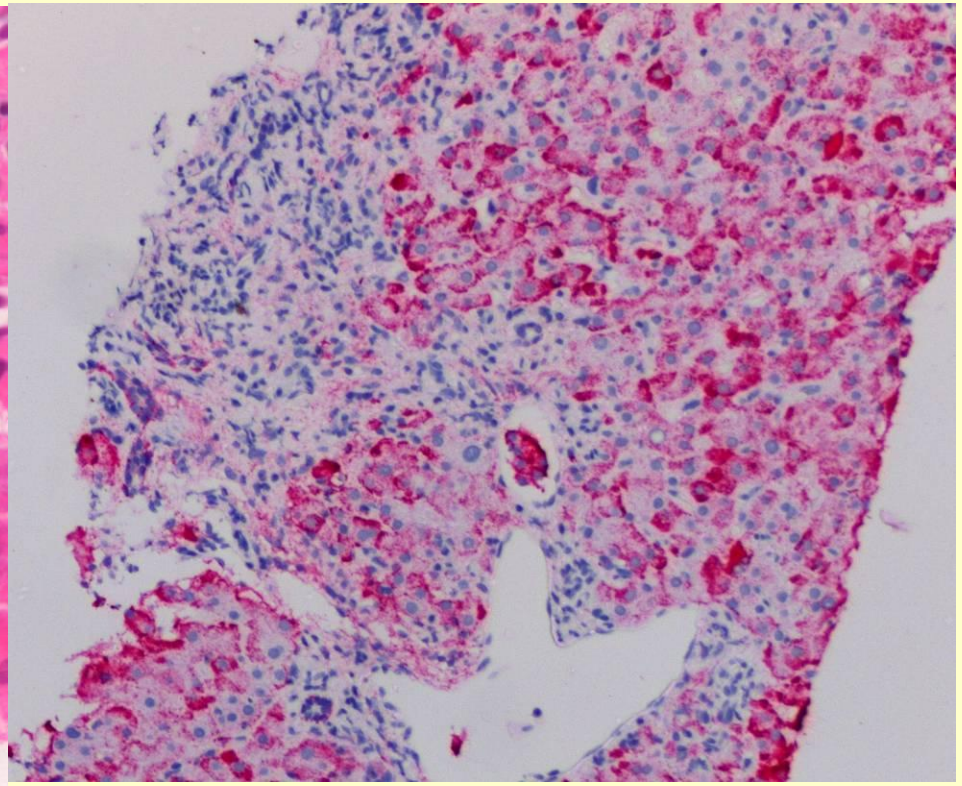
HBsAg

- HBsAg staining patterns
  - Scattered individual hepatocytes
    - High HBV DNA levels
  - Clusters of cells
    - Low HBV DNA levels
- Antiviral agents that inhibit HBV DNA polymerase and effectively reduce serum HBV DNA may not affect HBsAg production
  - Production of HBsAg by residual intrahepatic cccDNA is independent of viral replication
  - HBsAg seroclearance is associated with favourable biochemical, virological and histological parameters
- Elimination of intrahepatic HBs could indicate a favourable prognosis ie “cure” and permit cessation of nucleos/tide treatment



HBV flares vs autoimmune hepatitis



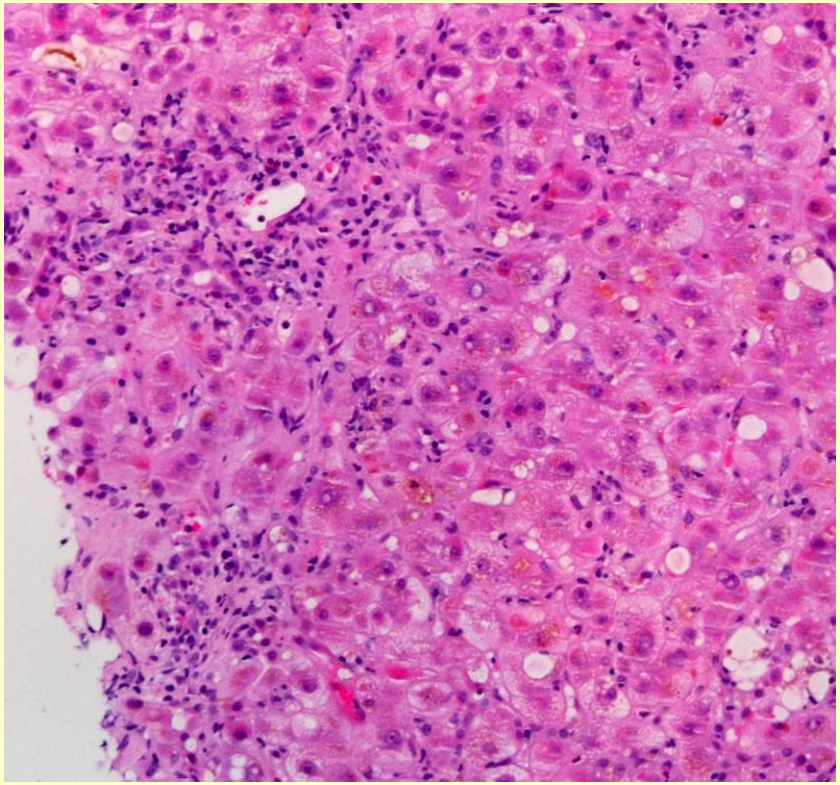


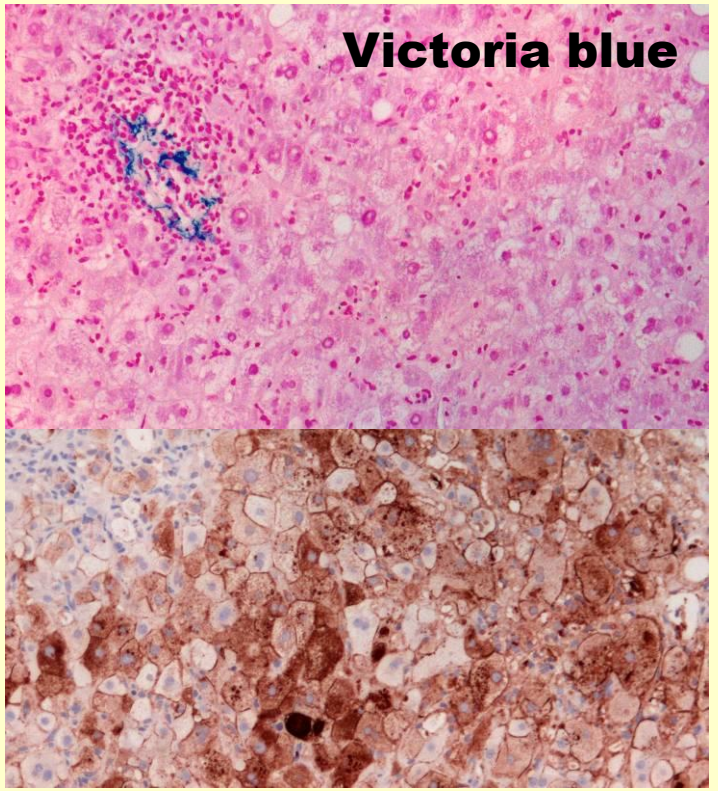
HBV during flare with plasma cells at interface:
HBsAg immunostaining can be informative
NB: HBV reactivation with chemotherapy / immunosuppression;
also immune reconstitution flare with tapering immunosuppression

“Acute” hepatitis with HBV

HBsAg immunostaining can be informative

and is more sensitive than histochemistry

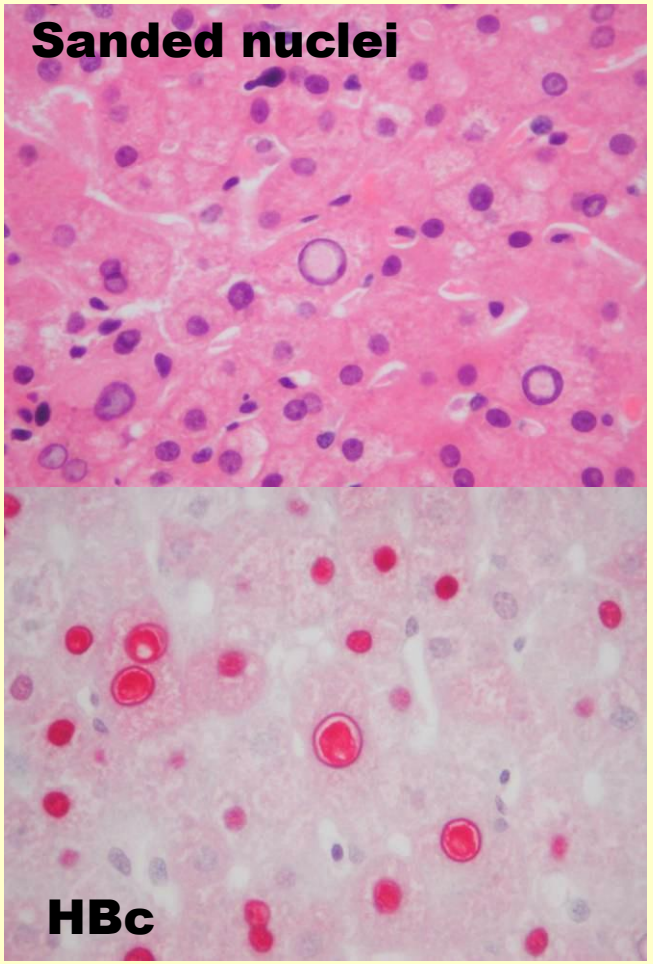




Morphologically acute hepatitis:
HBsAg immunostaining indicates chronic infection
Cholestasis due to incidental drug reaction
(causing “acute” presentation)

HBc

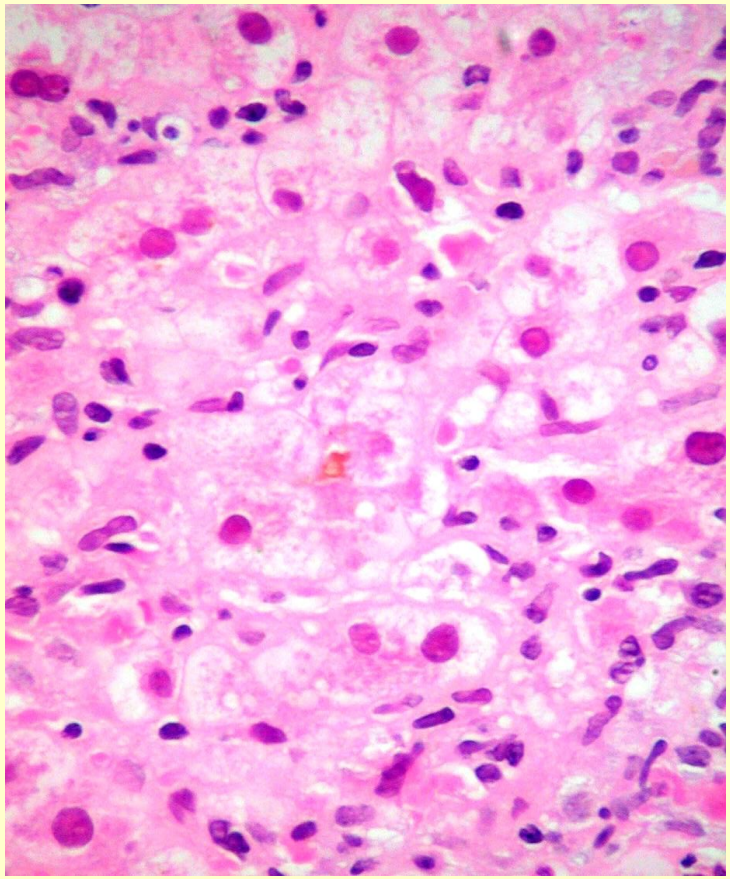
- **HBcAg in the liver correlates with viral replication, serum HBV DNA, and HBeAg**



Additional and alternative conditions are encountered in patients with chronic HBV hepatitis:

an important consideration when trying to base treatment decisions solely on "non-invasive" assessments such as serum HBVDNA and transaminases

- **Alcohol**
- **Drug reaction**
- **Infections:**
  - **HAV, HCV, HDV, HIV, tuberculosis**
- **Tumours:**
  - **Hepatocellular carcinoma**
  - **Cholangiocarcinoma lymphoma metastasis**



Cholestatic drug reaction in chronic HBV infection

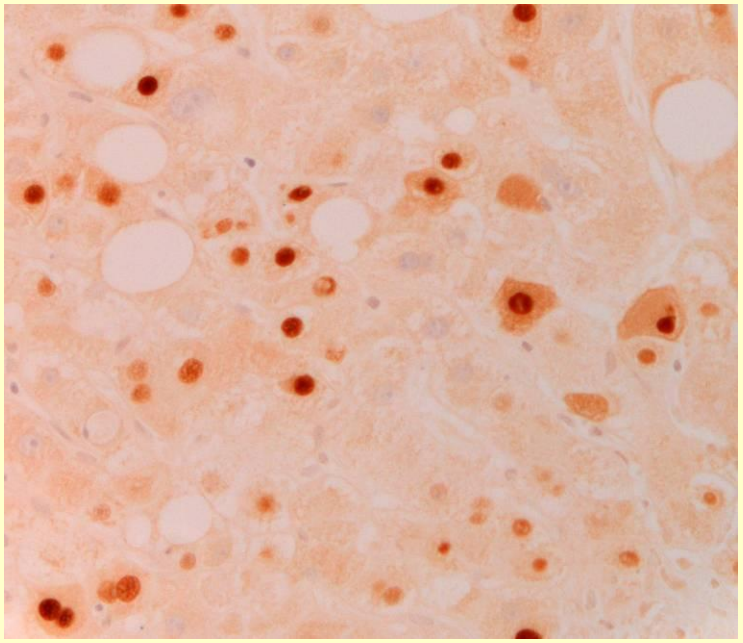
HDV

- **36nm single stranded RNA Deltavirus enveloped with HBsAg**
- **HDV/HBV co-infection or superinfection tend to increase hepatitis activity and chronicity compared to HBV alone**
- **Decline in HDV prevalence in Europe since first description of HDV by Rizzetto et al. in 1977 attributed to:**
  - **Improved socioeconomic conditions**
  - **“Universal” HBV vaccination**
    - **(except UK)**
  - **Measures introduced to control HIV**

HDV in London

Cross et al. J Med Virol 80,277;2008

- **Prevalence in South London 2000-2006 was 8.5% of HBV patients**
  - **HDV Ab screening of HBs Ag positive patients**
    - **HDV RNA not routinely tested**
- **Most HDV-infected subjects were born in regions where HDV is endemic:**
  - **Southern/Eastern Europe 28.1%**
  - **Africa 26.8%**
  - **Middle-East 7.3%**
- **Transmission**
  - **50% intra-familial**
  - **24.4% intravenous drug use**



HDV

Hepatitis B and D Conclusion

- **Duties of the pathologist regarding an HBV/HDV liver biopsy**
  - **Comment on the adequacy of the biopsy**
  - **State if the biopsy findings are diagnostic of, or consistent with chronic HBV infection**
  - **State the severity of inflammation and the stage of disease according to an agreed system (not necessarily a “scoring system”)**
  - **Make a comparison with previous liver biopsies**
  - **Indicate the existence of, or the possibility of additional conditions that may require further consideration clinically**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