

9th Bryan Warren School of Pathology
Pancreatic and Liver Pathology

Sarajevo, 6th -7th November 2015

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A Brief Introduction to the Liver Sessions

Lectures – Medical Liver Diseases:

1. How to approach a medical liver biopsy
2. Acute hepatitis (including acute liver failure)
3. Chronic hepatitis (viral and autoimmune)
4. Chronic biliary disease
5. Fatty liver disease

Slide Seminars:

- Focal liver lesions – hepatocellular and biliary/other

A Brief Introduction to the Liver Sessions

Main Objectives

1. Practical diagnostic issues – role of liver biopsy in the diagnosis and management of liver disease
2. Applications and limitations of liver biopsy
3. Recent developments and areas where there diagnostic difficulties and pitfalls
4. An approach to focal hepatic lesions

A Brief Introduction to the Liver Sessions

Other Points:

1. Audience participation

- Please ask questions!

2. Additional teaching material

- Handout on “*An Approach to Medical Liver Biopsies*” can be made available – either via course organisers or e mail – s.g.hubscher@bham.ac.uk

How to Approach a Medical Liver Biopsy

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Assessment of Medical Liver Biopsies

1. Role of liver biopsy in management of patients with medical liver diseases

- Utility of liver biopsy for diagnosis and management
- Limitations of liver biopsy
- Changing role of liver biopsy

2. Importance of recognising patterns of liver injury

3. Putting histological findings into clinical context

- Importance of clinico-pathological correlation

4. Areas where diagnostic problems frequently occur

Liver Biopsy Assessment Diagnostic Considerations

Liver Biopsy Assessment – Diagnostic Considerations

1. What information is required clinically?
2. Is the sample adequate?
3. Is the biopsy representative?

Liver Biopsy – Indications & Diagnostic Role

- To establish/confirm a primary diagnosis
 - To provide additional information in cases where a primary diagnosis has already been established by other investigations
- e.g. Chronic viral hepatitis (HBV/HCV), autoimmune hepatitis
- Biopsy taken to assess disease severity (grade of inflammatory activity and stage of fibrosis), which may determine treatment options
- BUT**
- Increasing use of non-invasive markers of fibrosis (serum markers, FibroScan) is reducing the frequency of liver biopsy to stage fibrosis in chronic liver disease (e.g. HCV, NAFLD)

Indications for Liver Biopsy

Identifying/confirming the cause of liver disease

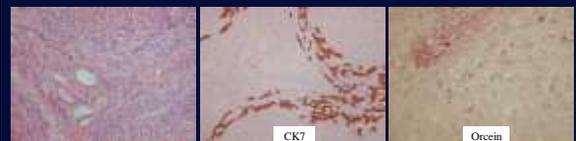
- (i) Cause of liver disease unknown**
 - e.g. Persistently abnormal LFTs - routine screening tests (viruses, autoantibodies etc) negative
- (ii) Cause of liver disease suspected.**
 - e.g. Autoimmune hepatitis
 - Presence of features supporting a diagnosis of AIH (e.g. plasma- cell rich portal inflammation, interface hepatitis, hepatocyte rosettes, emperipolesis)
 - Absence of features suggesting an alternative diagnosis (e.g. chronic biliary disease, fatty liver disease)
- (iii) More than one possible cause of liver disease suspected**
 - e.g. HCV and NAFLD
 - Liver biopsy may help to confirm dual pathology and identify the predominant cause of liver injury

Indications for Liver Biopsy

Identifying/confirming the cause of liver disease

- Importance of recognising patterns of liver injury (rather than specific diseases)
- Most of the common patterns of liver injury have several possible causes (e.g. acute hepatitis, chronic hepatitis, fatty liver disease, chronic biliary disease)
- Liver biopsy alone rarely diagnostic
- Clinico-pathological correlation essential

45 year old woman with pruritus and abnormal LFTs (ALT 2x normal, Alk Phos 3 x normal)



- Q1. What is the pattern of liver injury?
Q2. What are the most likely causes?

A1. Chronic biliary disease with bile duct loss
A2. PBC or PSC.

- Diagnostic duct lesions are present in < 50% of PBC liver biopsies and < 20% of PSC liver biopsies

(this woman had anti-mitochondrial antibodies and raised IgM)

Indications for Liver Biopsy

Importance of clinico-pathological correlation

- May be helpful to make an initial assessment of the biopsy without knowledge of the clinical details (to avoid subjective bias)
- Final interpretation should take into account all of the relevant clinical details and results of other investigations.
- Report should:
 - Include a summary of the clinical information /results of other investigations
 - Be structured in a way that addresses the main clinical question (s)

Liver Biopsy Assessment – Diagnostic Considerations

1. What information is required clinically?
2. Is the sample adequate?
3. Is the biopsy representative?

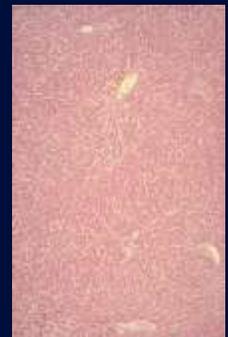
Liver Biopsy – Sampling Variability

- Average needle biopsy samples a tiny fraction (1/50-100,000th) of the whole liver
- Lesions which affect the liver diffusely can still be reliably assessed
 - e.g. fatty change, portal inflammation in chronic viral hepatitis
- Sampling variability may be a problem for diseases with an uneven distribution
 - e.g. fibrosis in chronic cholestatic diseases, bridging & pan-acinar necrosis in severe acute hepatitis
- Problems with sampling variability inversely proportional to the length and diameter of the biopsy specimen.

Primary Biliary Cirrhosis – Variable Fibrosis (similar changes in PSC & other chronic cholestatic disease)

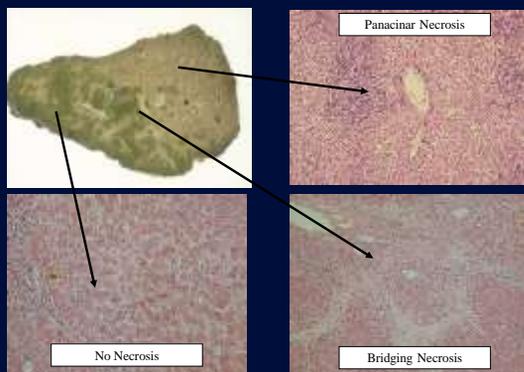


Cirrhotic area



No fibrosis

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



Changing Liver Biopsy Practice

Liver biopsies increasingly obtained by radiologists, who prefer to use thin needles (e.g. 18G or 20G)

Thin cores are suboptimal for assessing medical liver diseases:

- Incomplete/inadequate sampling of portal tracts
- Under-estimate disease severity (particularly fibrosis)

AASLD guidelines papers (Rockley 2009, Sanyal 2011) both recommend use of 16 gauge needles

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Recent AASLD guidelines papers (Rockley 2009, Sanyal 2011) both recommend use of 16 gauge needles

A comment on biopsy adequacy - length & width, fragmentation, number of portal tracts - may be appropriate as part of the pathology report (particularly if the material received is suboptimal / inadequate)

Histological Assessment of the Liver

Histological Assessment of the Liver – A Systematic Approach

	Normal Components	Abnormal Features
1. Architecture	<ul style="list-style-type: none"> • Vascular relationships 	<ul style="list-style-type: none"> • Fibrosis (severity and distribution) • Atrophy, nodular hyperplasia
2. Portal tracts	<ul style="list-style-type: none"> • Bile ducts • Hepatic arteries • Portal veins 	<ul style="list-style-type: none"> • Damage to/ loss of normal structures • Inflammation (severity & composition) • Interface hepatitis • Ductular reaction
3. Lobules	<ul style="list-style-type: none"> • Hepatocytes • Sinusoids • Hepatic veins 	<ul style="list-style-type: none"> • Damage to/ loss of normal structures • Kupffer cell enlargement/pigment deposits • Inflammation (severity & composition) • Sinusoidal dilatation / congestion

Other Investigations

4. Special stains	<ul style="list-style-type: none"> • Histochemical stains • Immunohistochemistry
5. Other investigations	<ul style="list-style-type: none"> • Electron microscopy • Other (e.g. biochemical/ molecular tests)

Assessment Of Liver Architecture

1. Are normal vascular relationships retained?
 - Distribution (e.g. periportal, perivenular, perisinusoidal)
 - Severity (staging)
 - Maturity
2. Is there any fibrosis?
 - Distribution (e.g. periportal, perivenular, perisinusoidal)
 - Severity (staging)
 - Maturity

Use Of Connective Tissue Stains In Liver Biopsy Assessment

Stain	Material Demonstrated	Distribution In Normal Liver
Reticulin	Type III collagen fibres	Portal tracts, hepatic sinusoids
Haematoxylin Van Gieson	Type I collagen fibres	Portal tracts, walls of hepatic veins
Orcein	Elastic fibres	Portal tracts, walls of hepatic veins

Connective Tissue Stains In Normal Liver



Use Of Connective Tissue Stains In Liver Biopsy Assessment

Stain	Material Demonstrated	Distribution In Normal Liver	Changes In Liver Disease
Reticulin	Type III collagen fibres	Portal tracts, hepatic sinusoids	Collapse of reticulin framework in areas of recent liver cell necrosis (days). Thickening of cell plates in areas of nodular regeneration
Haematoxylin Van Gieson	Type I collagen fibres	Portal tracts, walls of hepatic veins	Increased in hepatic fibrosis (weeks)
Orcein	Elastic fibres	Portal tracts, walls of hepatic veins	Found in long-standing fibrosis/cirrhosis (months/years)

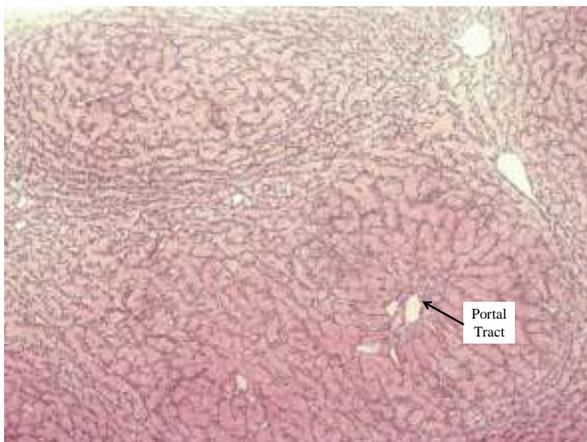
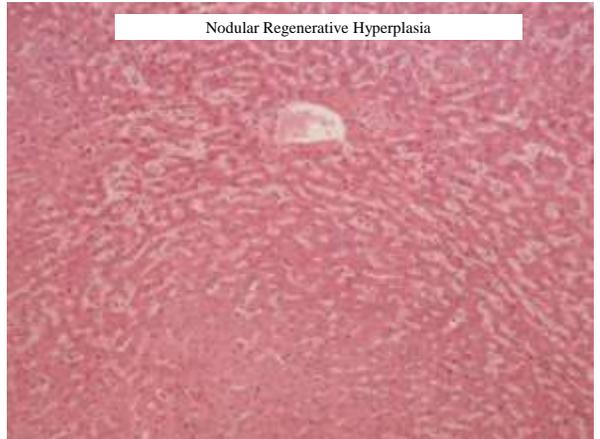
Assessment Of Liver Architecture

1. Are normal vascular relationships retained?
2. Is there any fibrosis?
 - Distribution (e.g. periportal, perivenular, perisinusoidal)
 - Severity (staging)
 - Maturity
3. Conditions in which assessment of architecture may be problematic:
 - Macronodular cirrhosis
 - Subtle changes in portal venous insufficiency (atrophy, nodular regenerative hyperplasia)
 - Nodular changes in recent post-necrotic collapse (see lecture on "Acute Hepatitis")

Fragmented Liver Biopsy in Cirrhosis
(glycogen storage disease)



Nodular Regenerative Hyperplasia



"Idiopathic" Non-Cirrhotic Portal Hypertension Problems with Liver Biopsy Diagnosis

- (1) Uncommon (compared with hepatic fibrosis/cirrhosis)
- (2) Histological changes subtle and patchy in distribution
- (3) Spectrum of morphological changes
 - Relationship between changes seen poorly understood
 - Confusion about terminology

"Idiopathic" Non- Cirrhotic Portal Hypertension

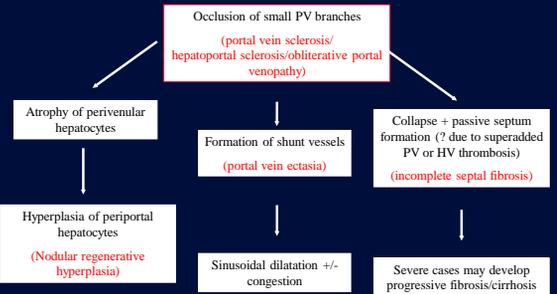
Definition

1. Evidence of portal hypertension (e.g. varices, splenomegaly)
2. Patent portal and hepatic veins
3. No cirrhosis (or significant fibrosis) on liver biopsy
4. No risk factors for chronic liver disease (e.g. alcohol, viral hepatitis)

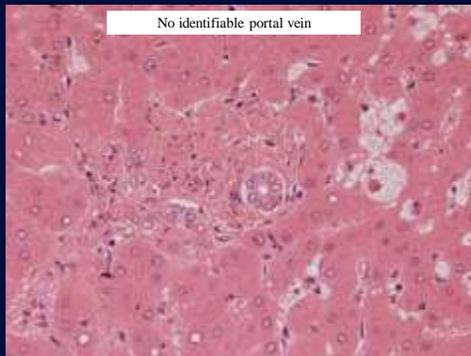
Alternative Terms

- Idiopathic portal hypertension (Japan)
- Non-cirrhotic portal hypertension
- Non-cirrhotic intrahepatic portal hypertension
- Hepatoportal sclerosis
- Non-cirrhotic portal fibrosis (India)
- Obliterative portal venopathy (Nayak 1979)

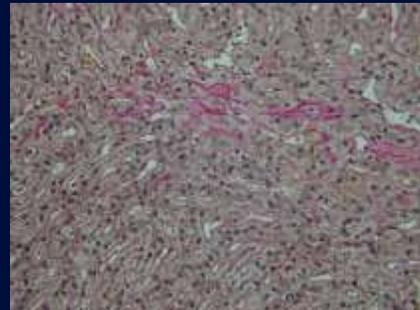
"Idiopathic" Non- Cirrhotic Portal Hypertension (hepatoportal sclerosis)
Pathogenesis + Histological Features



"Idiopathic" Non-Cirrhotic Portal Hypertension
Portal Tract Sclerosis (Obliterative Portal Venopathy)



Nodular Regenerative Hyperplasia

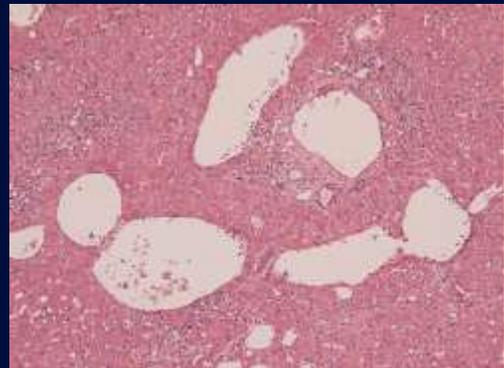


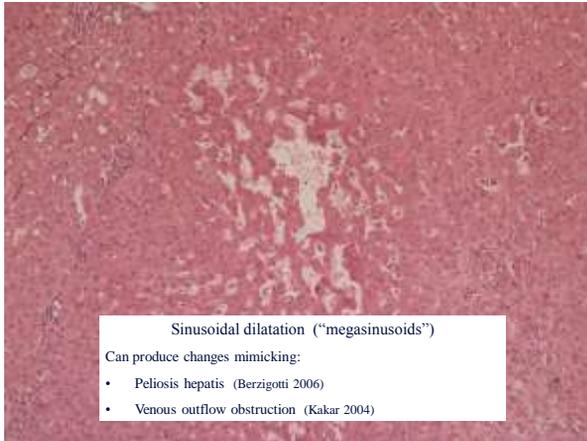
Sinusoidal Compression and Fibrosis at Periphery of Hyperplastic Nodules
• Further reduces sinusoidal blood flow, aggravating portal hypertension

"Idiopathic" Non-Cirrhotic Portal Hypertension
atrophy and septal fibrosis



"Idiopathic" Non-Cirrhotic Portal Hypertension
Periportal and Intralobular "Shunt vessels"





Histological Assessment of Liver Disease

Main Patterns of Liver Damage

1. Inflammation (portal, lobular, mixed)
2. Biliary features (bile duct damage, ductular reaction)
3. Hepatocellular damage
 - cell death (apoptosis, necrosis)
 - other degenerative changes (e.g. fat, cholestasis)
4. Fibrosis (may progress to cirrhosis)

Histological Assessment of Liver Disease

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Patterns of Inflammation in the Liver

Portal Inflammation

- Most chronic liver diseases (e.g. viral, autoimmune)
- Also seen in acute hepatitis

Lobular Inflammation

- Main pattern in acute hepatitis
- Varying degrees of lobular inflammation also commonly present in chronic viral and autoimmune hepatitis
- Predominant pattern in some chronic liver diseases (e.g. fatty liver disease)

Mixed portal and lobular inflammation

Pattern of inflammation cannot reliably distinguish acute from chronic hepatitis

- Clinical context
- Assessment of fibrosis

Normal Liver

Small numbers of lymphocytes seen in normal portal tracts

- Distinguishing normal from pathological portal lymphocytosis can be difficult
- Problems with scoring portal inflammation (normal liver = Ishak grade 1)

Similar problems may apply to assessing lobular lymphocytes

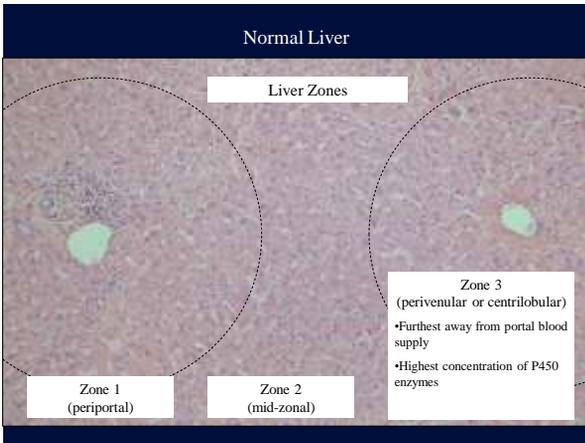
Normal Liver

Liver Zones

Zone 1 (periportal)

Zone 2 (mid-zonal)

Zone 3 (perivenular or centrilobular)



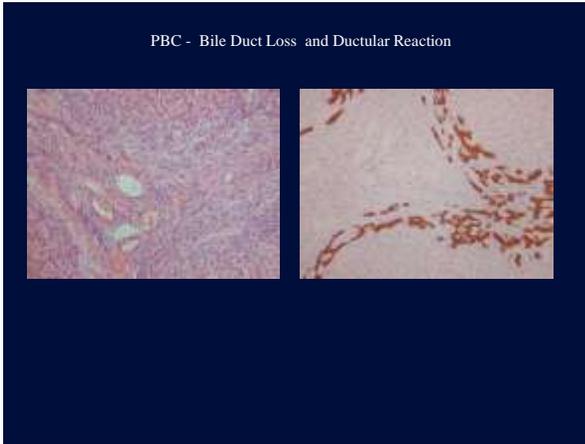
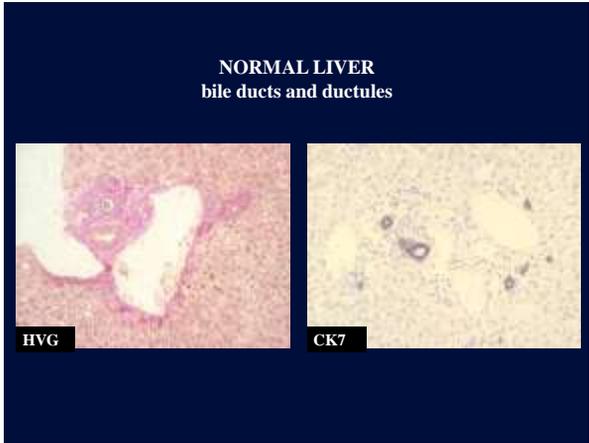
Histological Assessment of Liver Disease

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Interlobular Bile Ducts and Bile Ductules

	INTERLOBULAR BILE DUCTS	BILE DUCTULES
LOCATION IN NORMAL LIVER	CENTRAL (near hepatic arteriole)	PERIPHERAL (near canaliculi/canal of Hering)
SIZE	20-100µm	<20µm
MORPHOLOGY	Cuboidal/columnar epithelium. Clearly identifiable round lumen	Low cuboidal epithelium. Inconspicuous slit-like lumen
CHANGES IN LIVER DISEASE	Targets for damage (immune-mediated, ischaemic etc)	Ductular reaction (increase in number in response to bile duct damage – and other causes of liver disease)



Diseases Associated with Bile Duct Loss

1. Developmental
2. Immune-mediated
3. Vascular
4. Infective
5. Drugs/toxins
6. Neoplastic
7. 'Idiopathic'

Histological Assessment of Liver Disease

Main Patterns of Liver Damage

1. Inflammation (portal, lobular, mixed)
2. Biliary features (bile duct damage, ductular reaction)
3. Hepatocellular damage
 - cell death (apoptosis, necrosis)
 - Lecture on "Acute hepatitis"
 - other degenerative changes (e.g. fat, ballooning, cholestasis)
 - Lectures on "Chronic biliary disease" and "Fatty liver disease"
4. Fibrosis (may progress to cirrhosis)

Histological Assessment of Liver Disease

Main Patterns of Liver Damage

1. Inflammation (portal, lobular, mixed)
2. Biliary features (bile duct damage, ductular reaction)
3. Hepatocellular damage
 - cell death (apoptosis, necrosis)
 - other degenerative changes (e.g. fat, cholestasis)
4. Fibrosis (may progress to cirrhosis)

Patterns of Fibrosis in the Liver

Portal/periportal

- Most forms of chronic hepatitis (interface hepatitis)
- Chronic biliary diseases (ductular reaction)

Lobular (zone 3)

- Pericellular/perisinusoidal fibrosis (fatty liver disease)
- Confluent/bridging necrosis (e.g. autoimmune hepatitis)

Mixed (periportal and lobular)

- e.g. Fatty liver disease (particularly non-alcoholic)

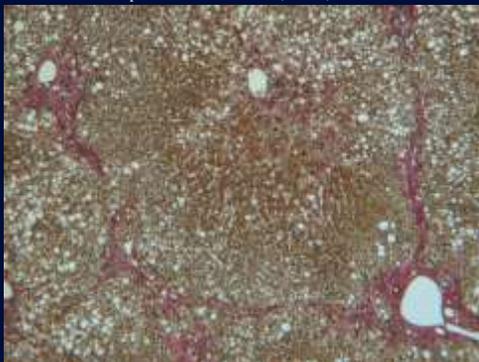


Periportal Fibrosis
(Autoimmune hepatitis)



Pericellular Fibrosis (zone 3)
(Non-alcoholic steatohepatitis)

Non-Alcoholic Steatohepatitis Periportal and Pericellular (zone 3) fibrosis



Chronic Liver Disease Assessing Disease Severity – Grading and Staging

Grading

- ongoing damage (usually inflammation) = "activity"
 - other features can be graded: e.g. fat, ballooning, hepatocyte apoptosis/necrosis
- potential to progress to chronic (irreversible) damage
- still potentially treatable

Staging

- progressive liver injury (usually fibrosis)
 - other features can be staged: e.g. bile duct loss
- less likely to be reversible

Semi-quantitative scoring systems

- Mainly used for hepatitis C, also in NAFLD
- Problems with observer variability, sampling variability, other limitations
- Use of non-invasive markers of liver fibrosis – liver biopsy less frequently used

Fibrosis Staging (Periportal Fibrosis)



Mild (fibrous portal expansion)



Moderate (bridging fibrosis)



Severe (cirrhosis)

Cirrhosis - Definition

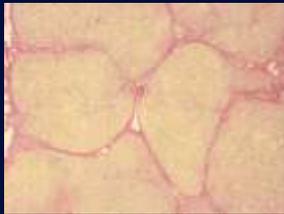
Irreversible (end-stage) condition affecting whole liver characterised by:

1. Loss of normal lobular architecture
2. Nodular regeneration
3. Fibrous septa

NOTE:

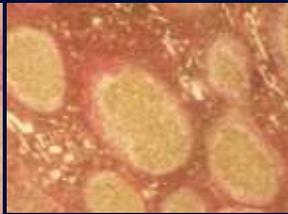
1. Fibrosis / early cirrhosis may be reversible
 - cases with thick septa /less surviving liver parenchyma (advanced cirrhosis) - less likely to be reversible, more likely to be associated with severe clinical signs (e.g. portal hypertension)
 - cirrhosis may be subclassified according to thickness of fibrous septa (e.g. Laennec system – stage 4a, 4b, 4c)
2. Uneven distribution of fibrosis/cirrhosis in some diseases (e.g. PBC & PSC)

Cirrhosis



Early Cirrhosis

- Thin septa
- Some preservation of architecture
- More functioning liver parenchyma



Late Cirrhosis

- Thick septa
- Loss of architecture
- Less functioning liver parenchyma

Morphometric Assessments – Surface Area Occupied by Collagen (“collagen proportionate area”)

(From Standish et al 2006)

- May be superior to conventional histological staging
 - More reproducible
 - Better predictors of outcome

Histological Assessment Of Medical Liver Biopsies Summary And Conclusion

1. Histological assessment of medical liver biopsies involves a systematic evaluation of histological abnormalities to identify one or more patterns of liver injury.
2. The final interpretation depends on correlating histological findings with clinical history and results of other relevant investigations
3. The information provided in the concluding section of the report should address the reason(s) indicated clinically for obtaining the biopsy.
4. The possibility of sampling variability should be considered if there is a disparity between the clinical and histological findings.
5. Scoring of histological features has limited value in routine clinical practice and should not be used as a substitute for conventional liver biopsy reporting.



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

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