Investigations for Liver Disorders

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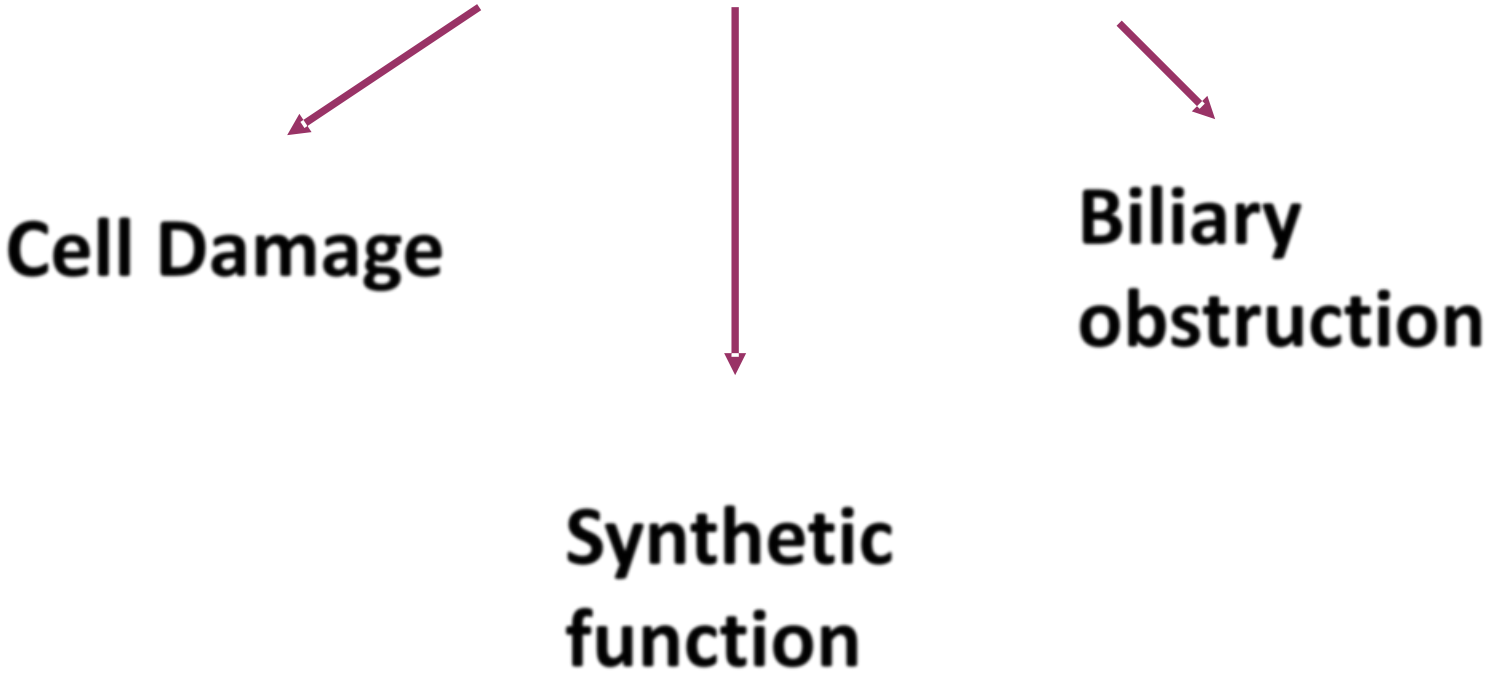
Learning Outcomes

- What are the common aetiological agents causing liver disorders?
- What are the common biochemical tests used in liver disorders?
- What are the types and causes of jaundice?
- What are the common metabolic disorders of the liver?
- What are the biochemical features of liver failure?
- What is the diagnostic approach to a patient with chronic liver disease?

What are the aetiological agents causing acute hepatitis?

- Infections
  - Hepatitis A,B,C,D,E
  - Cytomegalovirus
  - Epstein Barr virus
- Toxins & drugs
  - Alcohol
  - Paracetamol overdose
- Metabolic disorders
  - Inherited
  - Acquired
- Auto-immune disorders

Why are biochemical tests requested?



Biochemical Investigations

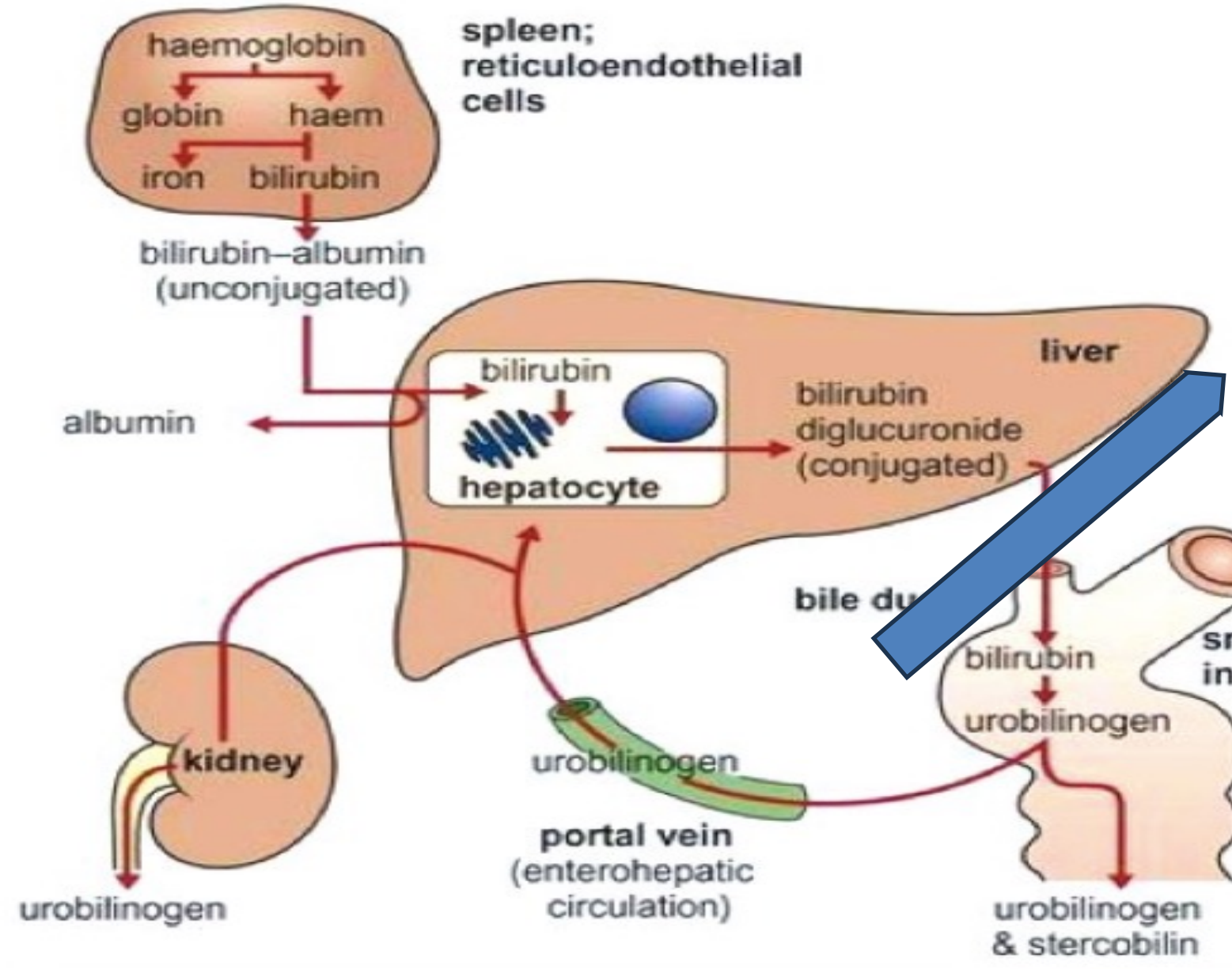
- Urine
  - Urine urobilinogen
  - Urine bilirubin
- Serum
  - Aminotransferases (ALT & AST)
  - Alkaline phosphatase
  - Gamma glutamyltransferase
  - Bilirubin & fractions

What are the tests used to assess the synthetic function of hepatocytes?

Serum albumin

Prothrombin time

Bilirubin Metabolism



Conjugated bilirubin reaches the systemic circulation

Colourless

Dark Brown

Jaundice

- Appears when serum bilirubin exceeds 50 µmol/L (3mg/dL)
- What are the causes in an adult?
  - Pre hepatic
  - Hepatic
  - Post hepatic

Causes of Jaundice

Pre hepatic	Hepatic	Post hepatic
Haemolysis Ineffective erythropoiesis	**Hepatitis** **Drugs,e.g rifampicin** **Gilbert syndrome**	Gallstones Biliary Stricture CA pancreas or biliary tree cholangitis

Serum Bilirubin

Tested by reaction with Diazo reagent
(Van den Bergh reaction)

Indirect reacting :

Unconjugated bilirubin

Direct reacting :

Conjugated bilirubin

Unconjugated bilirubin

Bound to albumin

Not filtered by renal glomeruli

Water insoluble

> 95% of total bilirubin normally

Conjugated bilirubin

- Water soluble
- Excreted in urine

Bilirubin is not detectable in urine normally,
bilirubinuria is always pathological

Normal serum total bilirubin concentration 0.2 - 1.2 mg/dL (3 - 20 µmol/L)
Jaundice is clinically apparent at a level of
> 2.5 mg/dL (> 50 µmol/L)

Urine Bilirubin

Only conjugated bilirubin is filtered by glomeruli

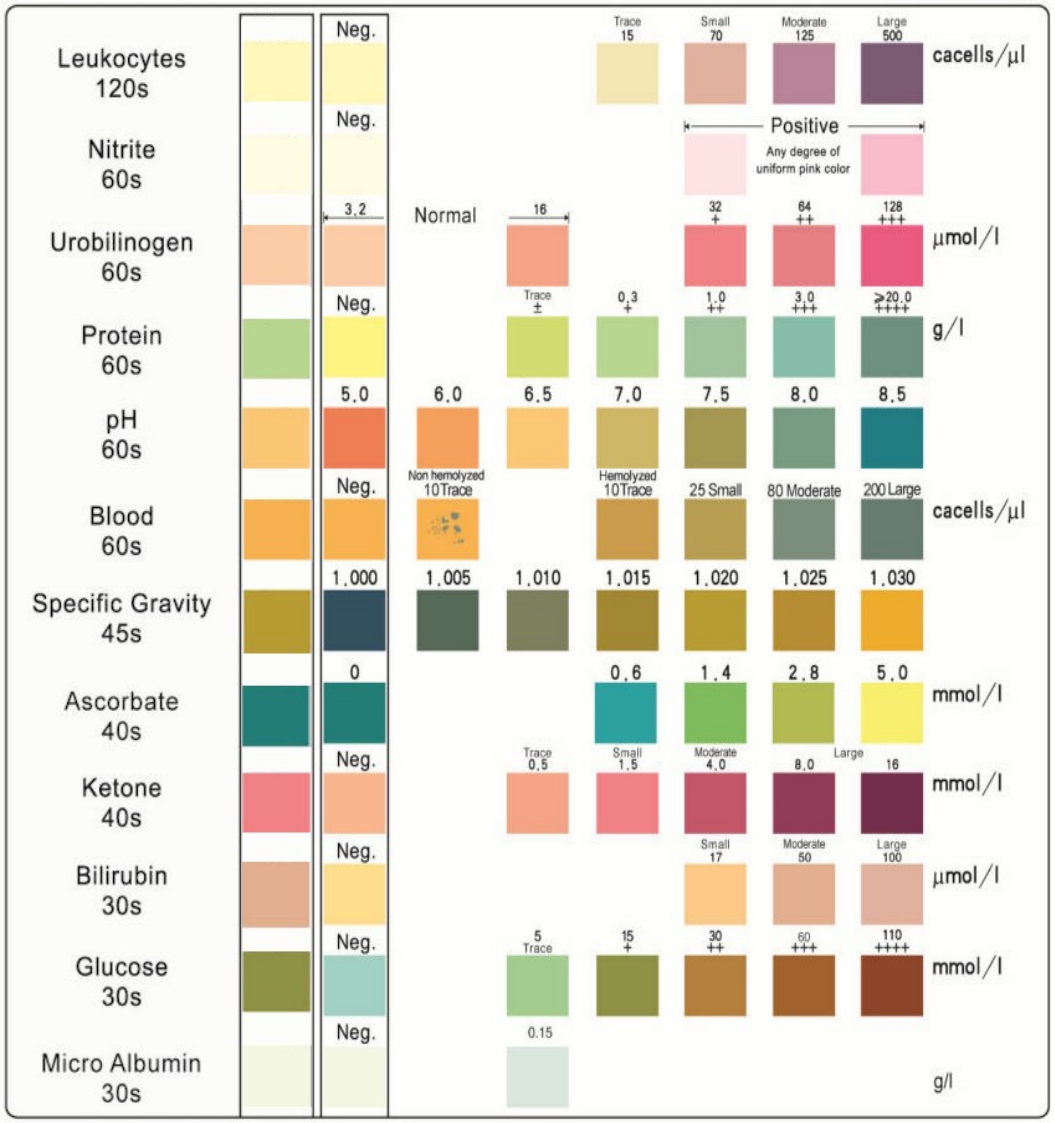
Fouchet's test

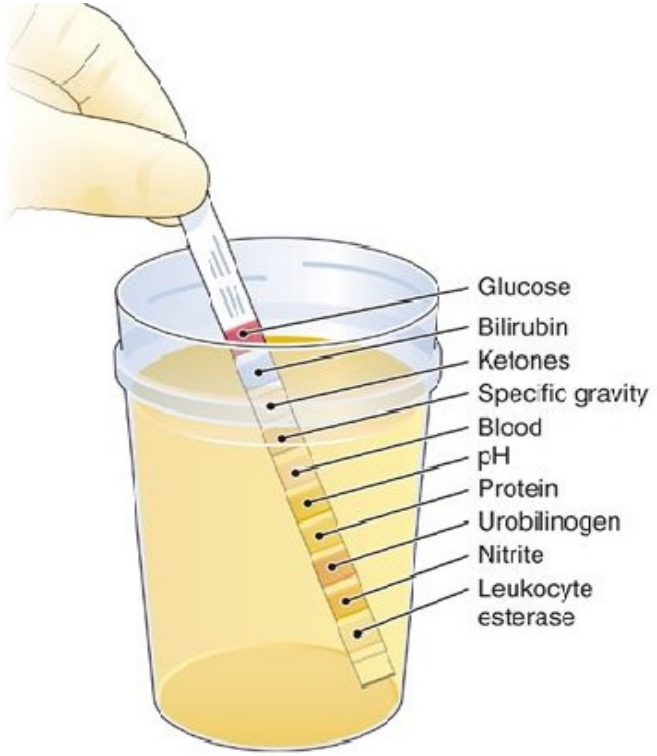
Greenish blue colour – positive result

Ictostix

Urinary test strips

Urine Strip


TESTING AND READING TIME
Rev. 08/2010
Leukocytes 120s
Nitrite 60s
Urobilinogen 60s
Protein 60s
pH 60s
Blood 60s
Specific Gravity 45s
Ascorbate 40s
Ketone 40s
Bilirubin 30s
Glucose 30s
Micro Albumin 30s
Neg.
3.2
5.0
1.000
0
Trace 15
Small 70
Moderate 125
Large 500
cacells/µl
Positive
Any degree of uniform pink color
Normal
16
32 +
64 ++
128 +++
µmol/l
Trace ±
0.3 +
1.0 ++
3.0 +++
≥20.0 ++++
g/l
6.0
6.5
7.0
7.5
8.0
8.5
Non hemolyzed 10 Trace
Hemolyzed 10 Trace
25 Small
80 Moderate
200 Large
cacells/µl
1.005
1.010
1.015
1.020
1.025
1.030
0.6
1.4
2.8
5.0
mmol/l
Trace 0.5
Small 1.5
Moderate 4.0
Large
8.0
16
mmol/l
Small 17
Moderate 50
Large 100
µmol/l
5 Trace
15 +
30 ++
60 +++
110 ++++
mmol/l
0.15
g/l


Glucose
Bilirubin
Ketones
Specific gravity
Blood
pH
Protein
Urobilinogen
Nitrite
Leukocyte esterase

Biochemistry of Jaundice

	Pre-hepatic	Hepatic	Post-hepatic/ cholestatic
Type of bilirubin elevated	Unconjugated bilirubin	Both conjugated and unconjugated bilirubin	Conjugated bilirubin
Urine urobilinogen	Increased	Decreased	Absent
Urine bilirubin (bile pigment)	Absent (acholuric)	++	+++
Urine bile salts	Absent	+	++
Urine colour	Normal	Dark	Dark
Stool colour	Dark brown	Normal/decreased	Clay colour

Neonatal Jaundice

- Physiological jaundice
- Haemolytic disease of the newborn
- Breast milk jaundice
- Hypothyroidism
- Neonatal hepatitis
- Metabolic disorders
- Biliary atresia

When to investigate neonatal jaundice

- Present at birth
- Appears within the first 24 hours of life
- Persisting > 14 days after birth
- Total serum bilirubin> 250 µmol/L
- Conjugated hyperbilirubinaemia
- Jaundice with other signs/symptoms of disease

Viral Hepatitis

Hepatitis A	Hepatitis B	Hepatitis C	Hepatitis D	Hepatitis E
Faeco-oral	Parenteral/sexual contact/perinatal	Parenteral	Parenteral	Faeco-oral
2-4 weeks	1-4 months	7–8	1-4 months	4–5 weeks
Never	10%	weeks ~80%	5% (co-infection); ≤70% for super-infection	Never
Anti-HAV IgM	HbSAg or Antibody for Core Ag	PCR for HCV RNA; 3rd-generation ELISA for antibody detection	Detection of IgM and IgG. antibodies; HDV RNA serum; HDAg in liver	PCR for HEV RNA; detection of serum IgM and IgG antibodies

Common features of viral hepatitis

- Cell injury
  - Ballooning
  - Apoptosis & necrosis
- Inflammation
  - Peri-portal
  - Lymphocytic infiltration
- Cholestasis

What are the expected changes in liver biochemistry in viral hepatitis?

- Markedly elevated transaminases
- Mild to moderate elevation of ALP & GGT
- Elevated total & conjugated bilirubin
- Presence of urinary bilirubin

What are the other causes of raised transaminases?

- Ischaemic hepatitis (following shock)
  - Sudden rapid rise in transaminases
- Acute drug or toxic induced liver injury
  - E.g.Paracetamol poisoning
- Acute exacerbation of chronic hepatitis B or autoimmune chronic active hepatitis
- Other systemic infections
  - Dengue

What are the causes of an isolated elevation of ALP?

- Space occupying lesions in the liver
  - Primary and secondary tumours
  - Abscesses
  - Cysts
- Primary biliary cirrhosis
- GGT is useful to differentiate between hepatic and non-hepatic causes of ALP

  elevation

Hepatotoxic drugs

- Acetaminophen (Paracetamol)
- Rifampicin
- Methyldopa
- Phenytoin
- Halothane
- Isoniazid
- Methotrexate
- High dose chemotherapy

Alcoholic Liver Disease

- Hepatic Steatosis
- Alcoholic hepatitis
- Cirrhosis
- Biochemical changes
  - Raised GGT
  - AST/ALT ratio >2
  - Raised triglycerides

Paracetamol Poisoning

- A single overdose of 10 g or a dose exceeding 200 mg/kg body weight

- Toxic metabolite is the NAPQI (N-acetyl-*p*-benzoquinoneimine)

- Glutathione detoxify this metabolite
- In overdose glutathione is depleted

- Toxic damage to hepatocytes from NAPQI & oxidative stress due to lack of glutathione

Paracetamol Poisoning –Clinical Features

- <24 hours
  - Anorexia, nausea and vomiting
- 24 –48 hours
  - Abdominal pain, tender hepatomegaly
- > 48 hours
  - Jaundice, encephalopathy, liver failure, acute kidney injury

Paracetamol Poisoning -Investigations

- Aminotransferases elevated
- Bilirubin elevated
- PT/INR prolonged (the best marker of severity)
- Poor prognosis if, after 24 hours

  Serum creatinine rises & acidosis develops

- Paracetamol levels should be checked between 4 -15 hours of ingestion
- N-acetylcysteine should be given based on the PCM level and time after ingestion

Features of Poor Outcome in Patients with PCM poisoning

- Acidosis (pH < 7.3)
- High INR (>6.5)
- Elevated serum creatinine
- Encephalopathy

Metabolic Liver Diseases

- Results from disordered metabolism
- Acquired or inherited
- The most common acquired disease
  - Nonalcoholic fatty liver disease (NAFLD)

Inherited Metabolic Diseases

- Wilson disease
- Haemochromatosis
- Alpha 1- antitrypsin deficiency

Alpha1- antitrypsin deficiency

Associated with

- Neonatal hepatitis
- Cirrhosis in infancy and childhood

Haemochromatosis

Haemochromatosis and other iron overload disorders associated with liver pathology are characterized by:

Increase ferritin
Increase iron saturation of transferrin e.g (> 80%)

Wilson Disease

- Inherited abnormality of Copper metabolism
- Decreased biliary excretion of Cu and incorporation to caeruloplasmin
- Biochemical features
  - Reduced plasma caeruloplasmin
  - Low/ low normal plasma Cu
  - Increased urinary Cu excretion

Non-alcoholic Fatty Liver Disease

- Hepatic manifestation of the metabolic syndrome
- Prevalence: worldwide 10 –24%
- Higher in obesity: 50 –75%
- The commonest cause for altered LFT after the exclusion of
  - Viral hepatitis
  - Alcoholism
  - Inherited liver disorders
  - Medications

NAFLD -Diagnosis

- Alcohol consumption < 20g/day
- Evidence for hepatic steatosis
  - Imaging
  - Liver biopsy

NAFLD

- Factors determining severity
  - Extent of fibrosis
  - Degree of inflammation
- Associated with
  - Type 2 DM
  - Hypertension
  - Obesity
  - Older age

NAFLD -Biochemistry

- Transaminases are neither sensitive nor specific
- May be elevated 2-4 times above the ULN
- GGT is often elevated
- **Liver biopsy is the confirmatory test**

Acute Liver Failure

- A range of clinical syndromes
- Severe liver dysfunction and encephalopathy coexist
- Develops within the first six months after the onset of acute liver disease
- Common causes
  - Viral hepatitis (except Hepatitis C)
  - Paracetamol poisoning

Biochemical Features

- Severe hyponatremia
- Hypocalcaemia
- Hypoglycaemia
- Low blood urea
- Prolonged prothrombin time

Cirrhosis

- No reliable biochemical tests for diagnosis
- Moderate or persistently elevated transaminases
- Liver biopsy is the confirmatory test
- Biochemical tests may help identify cause
  - Fasting transferrin saturation
  - Ceruloplasmin
  - Hepatitis B, C serology

An approach to diagnosis

- Simple biochemistry
- Cholestatic or hepatocellular
- True liver function tests
  - –Acute vs chronic
- Check for past liver biochemistry tests
- Imaging
- Further information
  - Alchohol, medications, herbal medicine

References

Marshall WJ, Bangert SK and Lapsley.Clinical chemistry 9th Edition

Tietz Textbook of Clinical Chemistry and Molecular Diagnostics – 5th Edition