• Overall plan and function of the liver
• Bilirubin physiology
• Understand bilirubin as biomarker for liver disease
• Bilirubin; liver
Liver Functions

- Bilirubin metabolism
- Protein Synthesis
  - Albumin
  - Coagulation factors (II, V, VII, IX, X)
- Bile Salt Metabolism
- Lipid Metabolism
- Glycogen storage and gluconeogenesis
- Drug metabolism/Xenobiotic transformation
Lobules

Central vein

Portal triad:
- artery
- vein
- bile duct
# Bile

<table>
<thead>
<tr>
<th>Component</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>H₂O</td>
<td>84%</td>
</tr>
<tr>
<td>Bile Salts</td>
<td>11.5%</td>
</tr>
<tr>
<td>Phosphatidyl Choline (lecithin)</td>
<td>3.0%</td>
</tr>
<tr>
<td>Bile pigments, protein, inorganic ions</td>
<td>1.0%</td>
</tr>
</tbody>
</table>
Bilirubin

- Breakdown product of heme compounds
- Neurotoxic in infants
  - Secondary to immature blood-brain barrier
- Bilirubin metabolism is used as a marker to localize the site of liver disease
Bilirubin - Source

- Breakdown product of hemoglobin from ineffective erythropoiesis and red blood cell senescence (80%)
  - Reticuloendothelial cells mainly in the spleen and liver represent the major sites of breakdown
  - Enhanced with increased RBC turnover as seen in the hemoglobinopathies (e.g. Sickle-cell disease)

- Other heme containing compounds (20%)
Fate of $^{14}$C-glycine, a precursor of heme that is metabolized to bilirubin
RBC breakdown

- Turnover of RBC’s in the spleen, liver, bone marrow, and lymph nodes
  - Reticuloendothelial cells are phagocytic
FIG. 6. Pathways of biliverdin production from heme by chain-opening and bilirubin production by reduction of biliverdin. (Reproduced from Odell GB. Neonatal Hyperbilirubinemia. New York: Grune and Stratton; 1980:4, with permission.)
Bilirubin is hydrophobic and thus insoluble in blood
  ◦ It is transported in blood bound to albumin
• The major plasma protein
  ◦ Contributes to the total oncotic pressure of blood.

• A general carrier for many hydrophobic compounds
  ◦ High capacity for bilirubin
  ◦ Reversible
  ◦ Binding of bilirubin can be compromised by competition from other hydrophobic compounds
Competition for Albumin Binding

- Drugs: sulfonamides, streptomycin, chloramphenicol, ampicillin, salicylates, diuretics, food additives
- Free fatty acids
Bilirubin - Hepatic Uptake

- Bilirubin is unloaded from albumin and transported into the hepatocyte
- ~30% is taken up with each pass through the liver
Intracellular transport is mediated by ligandin, a cytoplasmic protein.
Bilirubin is then conjugated to carbohydrate (glucoronyl moieties) that increases water solubility
  - UDP-glucurononyltransferase
Bilirubin mono- and diglucuronide
Bilirubin Monoglucurononide
Bilirubin Diglucuronide
Excretion of bilirubin

- C-MOAT transporter (MRP2)
  - Member of the mdr family
- Transport of conjugated bilirubin
  - The most sensitive step in bilirubin metabolism
  - Sensitive to estrogens, infections
A HEME METABOLISM

Heme

O² + CO →

Billiverdin

NADPH + H⁺

NAD⁺

Billirubin

BH⁺

Microbial enzymes

Urobilinogen

Kidney

Urobilin in urine
Bilirubin-glucuronide → Bilirubin → Urobilinogen → Stercobilin → Excretion in feces

Small intestine
Laboratory Assessment

- Total bilirubin
- Direct bilirubin = “conjugated bilirubin”
  - Represents that hydrophilic fraction of bilirubin that is more readily accessible to diazo dyes. To determine the total bilirubin, an accelerator is added to make all the bilirubin reactive with the dye.
- Calculated indirect bilirubin = unconjugated bilirubin
• 36 year old pregnant woman presents with acute right upper quadrant pain
  ◦ Total bilirubin = 8.2 (0.1-1.2)
  ◦ Direct bilirubin = 7.9
### Normal Values for Bile duct obstruction, Hemolytic anemia, and Liver Failure

<table>
<thead>
<tr>
<th></th>
<th>Normal Values</th>
<th>Bile duct obstruction</th>
<th>Hemolytic anemia</th>
<th>Liver Failure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total bilirubin</td>
<td>0.3-1.3 mg/dl</td>
<td>↑</td>
<td>↑</td>
<td>↑</td>
</tr>
<tr>
<td>Direct bilirubin</td>
<td>0.1-0.3 mg/dl</td>
<td>↑</td>
<td>nl</td>
<td>↑</td>
</tr>
<tr>
<td>Indirect bilirubin</td>
<td>nl</td>
<td></td>
<td>↑</td>
<td>↑</td>
</tr>
</tbody>
</table>
• 46 year old man with colon cancer and recently discovered liver metastasis
  ◦ Total bilirubin = 15.2 (0.1-1.2)
  ◦ Direct bilirubin = 2.3
• Prothrombin time - 15 sec (normal < 12)
2 - day newborn who is brought back to the hospital jaundiced

- Total bilirubin = 11.0 (0.1-1.2)
- Direct bilirubin = 0.3
Kernicterus

- Bilirubin encephalopathy
  - Neonates have an immature blood-brain barrier
- Deposition of unconjugated bilirubin in the basal ganglia and brainstem nuclei
  - Usually 21-50mg/dL
- Can result in death or permanent neurological defects
An 18 year old male presents to the local draft board for his physical exam.

He is slightly jaundiced.

- Total bilirubin = 3.2 (0.1-1.2)
- Direct bilirubin = 0.2
• His little brother accompanying him says he has been fasting for the last two days
Gilbert’s Syndrome

- Polymorphism in the promoter region affect the expression of UDP-glucorononyltransferase
- Patients with (TA)7 instead of (TA)6 have lower UDP-G activity
- Exhibit mild elevations of bilirubin that is exacerbated by fasting, stress, or illness.
Crigler-Najjar Syndrome - mutations in the UDP-glucurononyltransferase gene
- Type I: Autosomal recessive with complete absence of activity leading to death
- Type II: Partial expression with some activity

Dubin-Johnson Syndrome
- Defects in CMOAT (MRP2) resulting in an excretory defect. Results in pigmented livers
Liver Function Tests
Liver Function Tests

- Where is the problem?
  - Biliary tract
    - gallstones
    - cholangiocarcinoma
  - Hepatocyte
    - viral or alcoholic hepatitis
  - Mixed
    - hepatocellular carcinoma
Liver Function Tests

• Is this an acute or chronic disease?
  ◦ Half-life of liver derived proteins
  ◦ If the liver stopped functioning today, how long would it take to see an abnormality in the blood?
Test