HEMOGLOBIN

Learning Objectives:

1. List the steps in the biosynthesis of Hemoglobin
2. Describle the function of Hemoglobin
3. Describe the fate of Hemoglobin
4. List the Normal and Abnormal Hemoglobin
5. Discuss the types of Jaundice

Structure of Hemoglobin

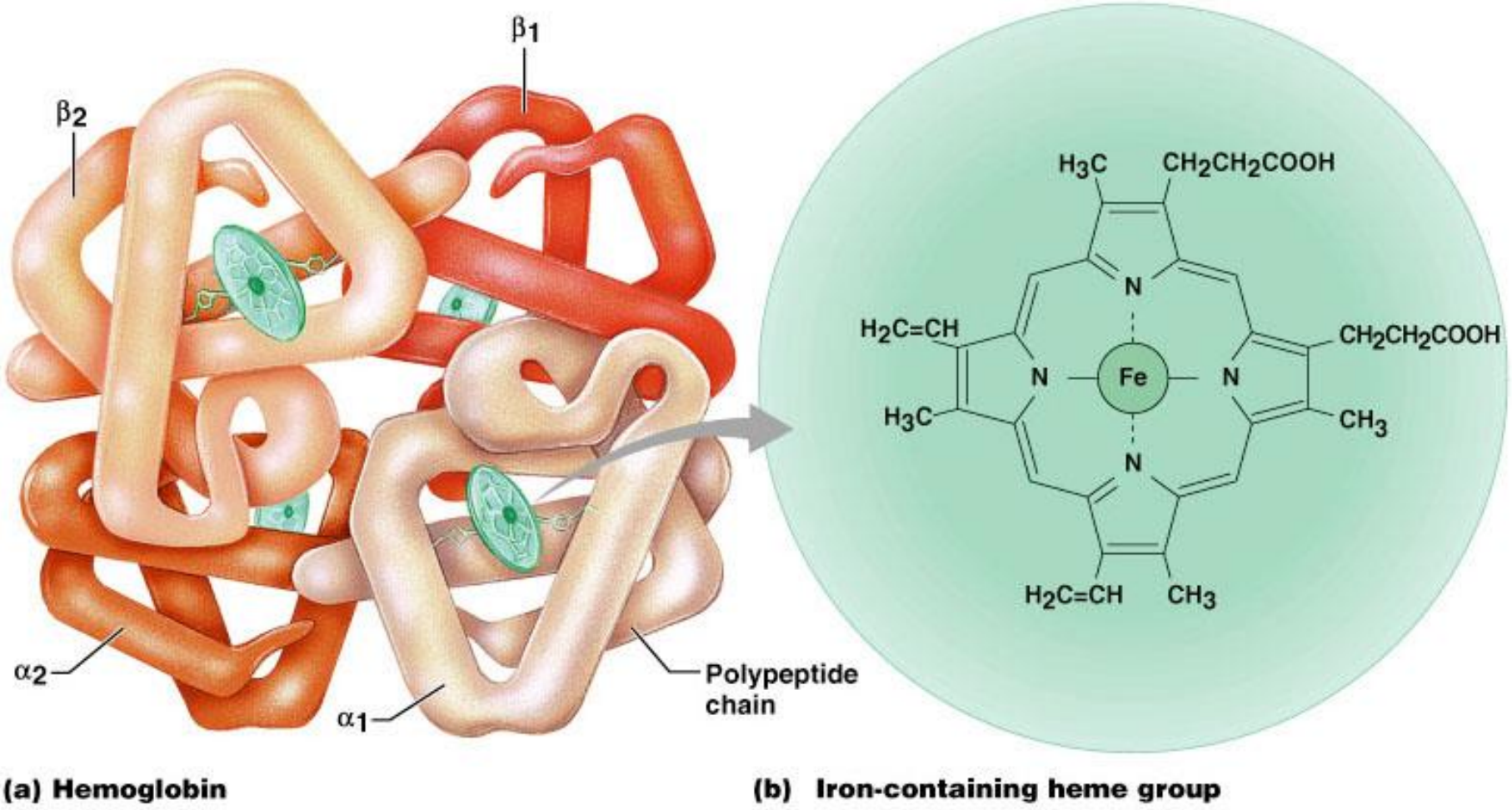


Figure 17.4

Hemoglobin

- Oxyhemoglobin – hemoglobin bound to oxygen
  - Oxygen loading takes place in the lungs
- Deoxyhemoglobin – hemoglobin after oxygen diffuses into tissues (reduced Hb)
- Carbaminohemoglobin – hemoglobin bound to carbon dioxide
  - Carbon dioxide loading takes place in the tissues

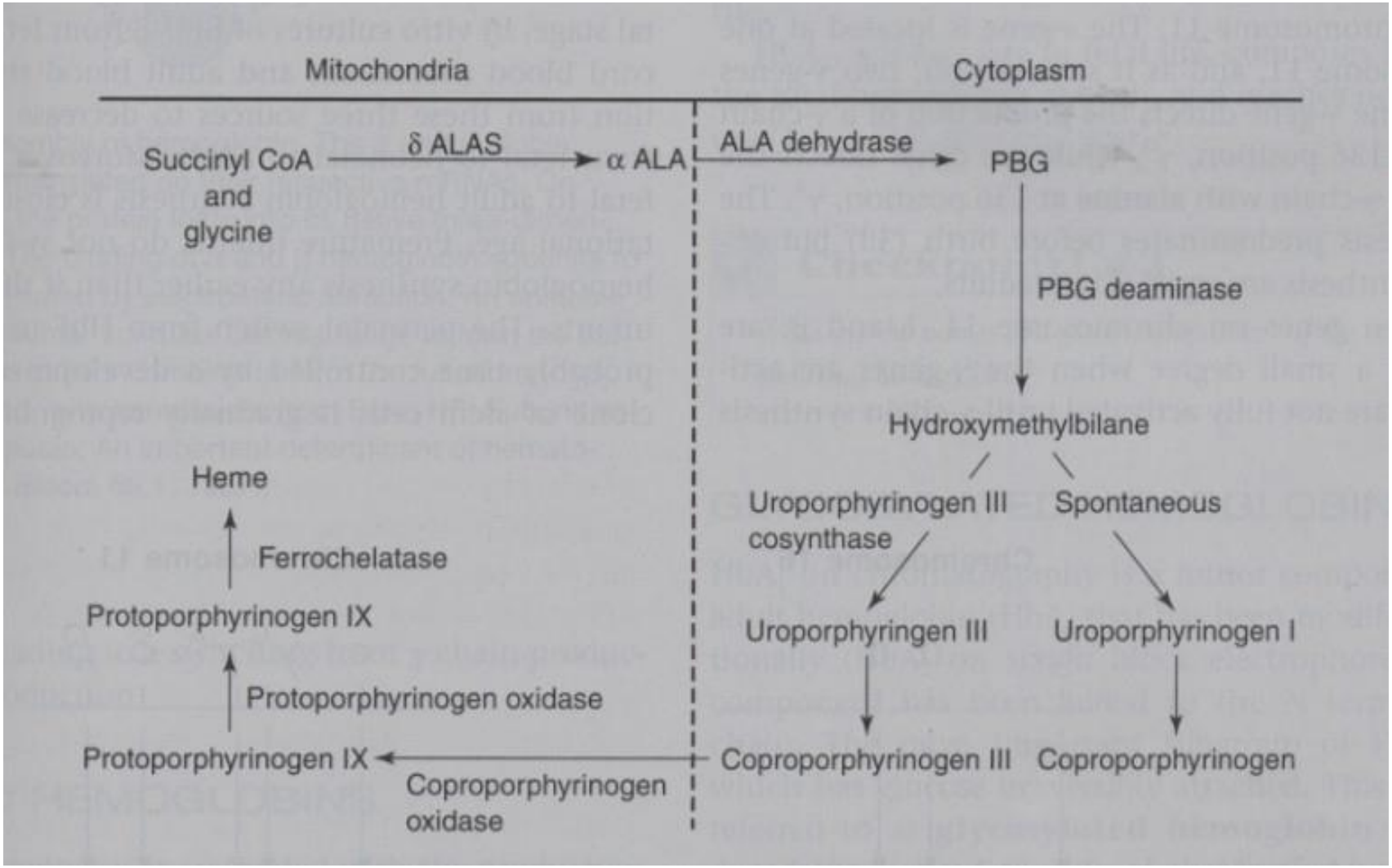
Synthesis of Haemoglobin (Hb)

- Haem & globin produced at two different sites in the cells
    - Haem in mitochondria
    - Globin in polyribosomes
- Well synchronized
- Normal hemoglobin production is dependent upon 3 processes: Adequate iron delivery and supply, adequate synthesis of protoporphyrins and adequate globin synthesis.

Hemoglobin Structure and Function

- Hemoglobin occupies 33% of the RBC volume and 90-95% of the dry weight.
  - 65% of the hemoglobin synthesis occurs in the nucleated stages of RBC maturation and 35% during the reticulocyte stage.
  - Normal hemoglobin consists of 4 heme groups which contain a protoporphyrin ring plus iron and globin which is a tetramer of 2 pairs of polypeptide chains.

Hemoglobin synthesis



Porphyria

- Since porphyrinogens are readily oxidized to form porphyrins, excess formation of porphyrins can occur if any of the normal enzymatic steps in heme synthesis is blocked.
- Inherited

  Erythropoietic porphyria - results from excessive production of porphyrins in the bone marrow.

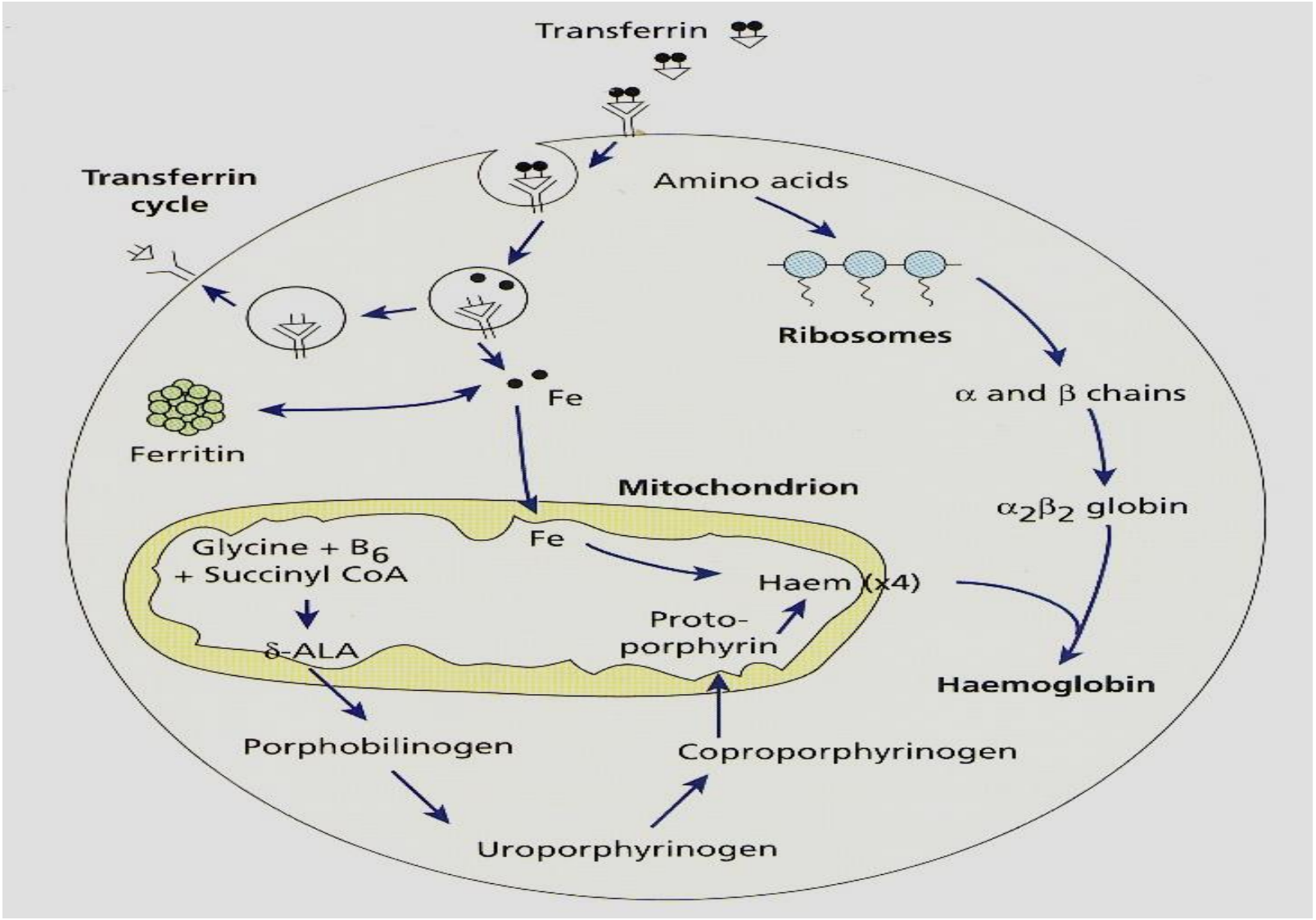
  Hepatic porphyria - results from excessive production of porphyrins in the liver.

  - Acquired

  Lead intoxication - interferes with protoporphyrin synthesis

  Chronic alcoholic liver disease

Synthesis of Haemoglobin



- Globin Synthesis
  - In the fetus and the adult 4 types of hemoglobin chains may be formed: alpha ( ), beta ( ), gamma ( ), and delta ( ).
  - Normal hemoglobin's contain 4 globin chains.
  - Hemoglobin (hgb) F= $_2$ $_2$ and is the predominant hgb formed during liver and bone marrow erythropoiesis in the fetus.
  - A normal, full term baby has 50-85% hgbF.
  - Near the end of the first year of life, normal adult hgb levels are reached.

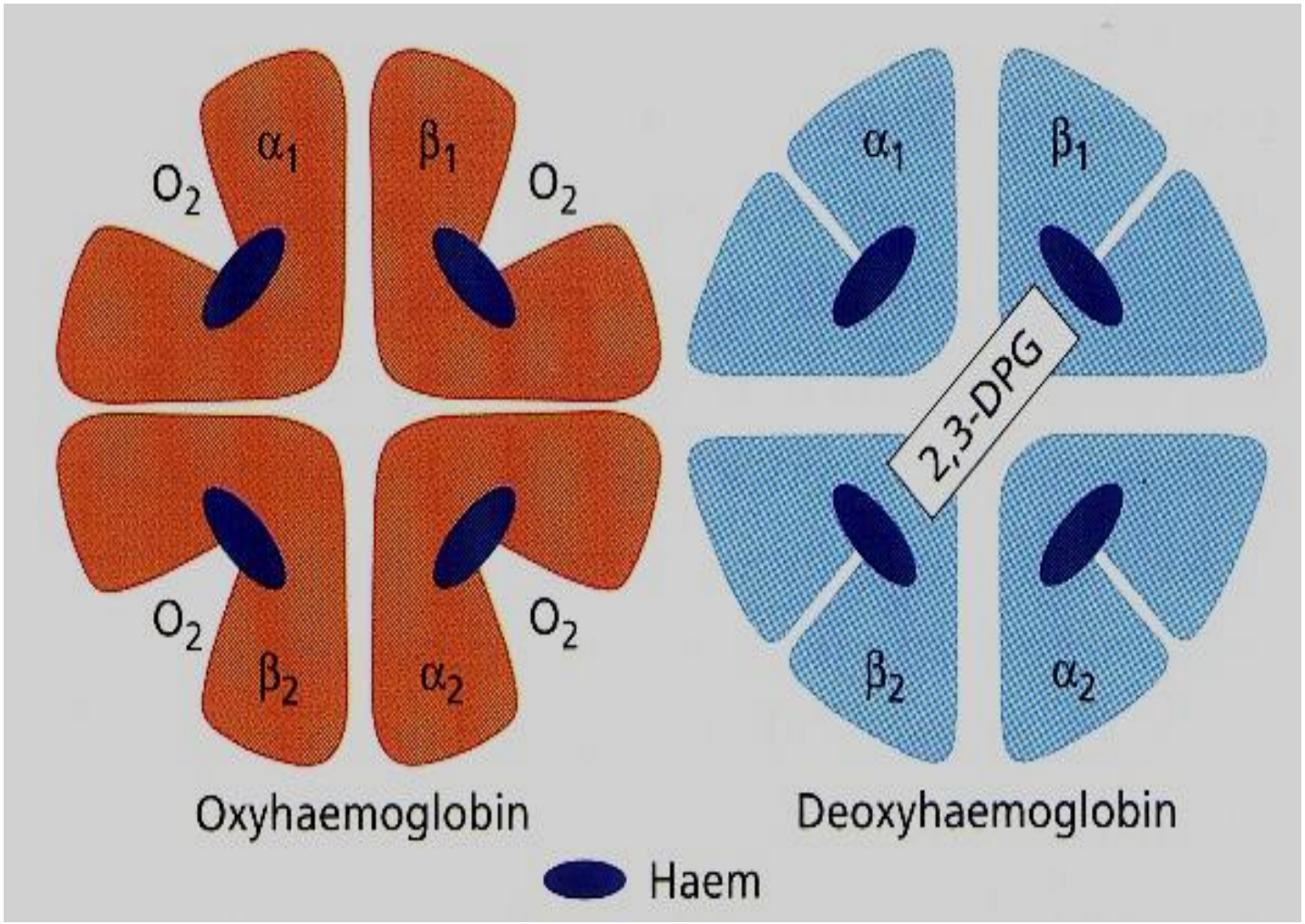
Hemoglobin Structure and Function

- Normal adult RBCs contain:
  - » 95-97% hgb A=$\alpha_2\beta_2$
  - » 2-3% hgb A2= $\alpha_2\delta_2$
  - » 1-2% hgb F (fetal hgb)= $\alpha_2\gamma_2$
  - – Each globin chain links with heme to form hgb= 4 globin + 4 heme.
  - – An adequate amount of globin synthesis is also important. A decreased production in 1 chain results in thalassemia.

Hemoglobin Structure and Function

- The primary function of hgb is gas transport.
  - In unloading the space between the chains widens and 2,3 diphosphoglycerate (DPG) binds. This is the T (tense) form of hgb and it is called deoxyhgb. It has a lower affinity for $O_{2,}$ so O_2 unloads from the hbg.
  - When hgb loads O_2 and becomes oxyhgb the chains are pulled together, expelling 2,3 DPG. This is the R (relaxed) form of hgb. It has a higher affinity for $O_{2,}$ so O_2 binds to or loads onto the hgb.

Oxy & deoxyhaemoglobin



Hemoglobin Structure and Function

- Acquired abnormal hgbs of clinical importance are those that have been altered post- translationally to produce hgbs that are unable to transport or deliver O_2 and they include:
  - Carboxyhgb - CO replaces O_2 and binds 200X tighter than O_2.
    - » This may be seen with heavy smokers
  - Methgb - occurs when iron is oxidized to the +3 (ferric) state. In order for hgb to carry O_2 the iron must be in the +2 (ferrous) state. In the body, normally~ 2% is formed and reducing systems prevent an increase beyond 2%.
    - » Increases above 2% can occur with the ingestion of strong oxidant drugs or
    - » As a result of enzyme deficiency.

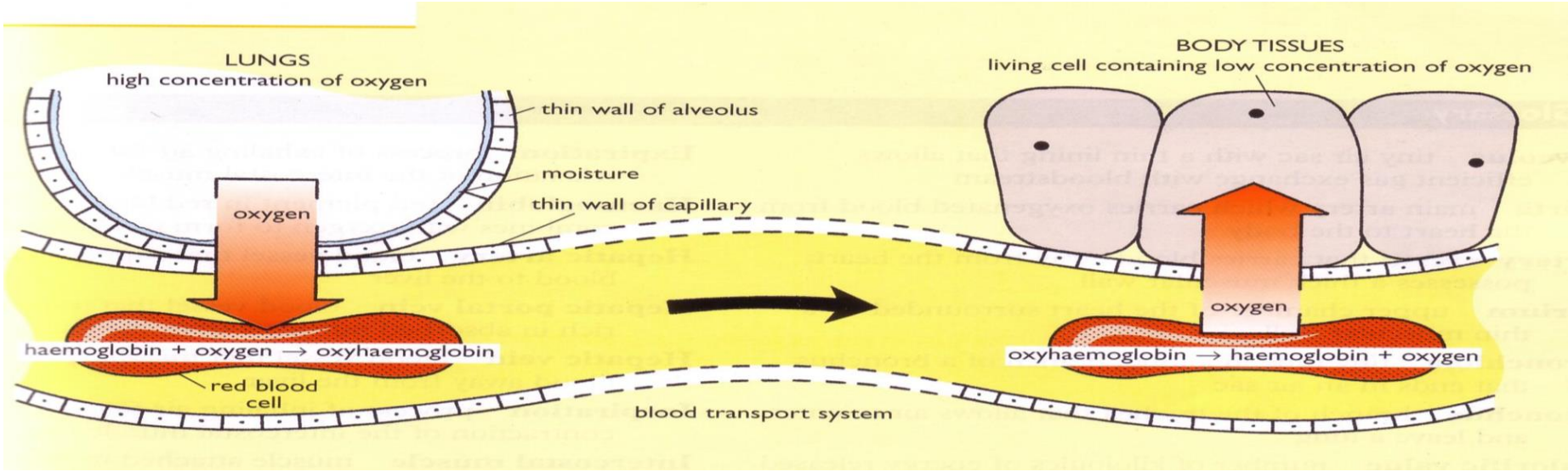
Hemoglobin Structure and Function

  - » Methgb can be reduced by treatment with methylene blue or ascorbic acid.
- – Sulfhgb - occurs when the sulfur content of the blood increases due to ingestion of sulfur containing drugs or to chronic constipation. Unlike 1 and 2 this is an irreversible change of hgb.

Erythrocyte destruction

- RBC destruction is normally the result of senescsence.
  - Each day ~ 1% of the RBCs are removed and replaced.
  - RBC aging is characterized by decreased glycolytic enzyme activity which leads to decreased energy production and subsequent loss of deformability and membrane integrity.
  - 90% of aged RBC destruction is extravascular and occurs mainly in the phagocytic cells in the spleen, with a small amount occurring in the liver and bone marrow.

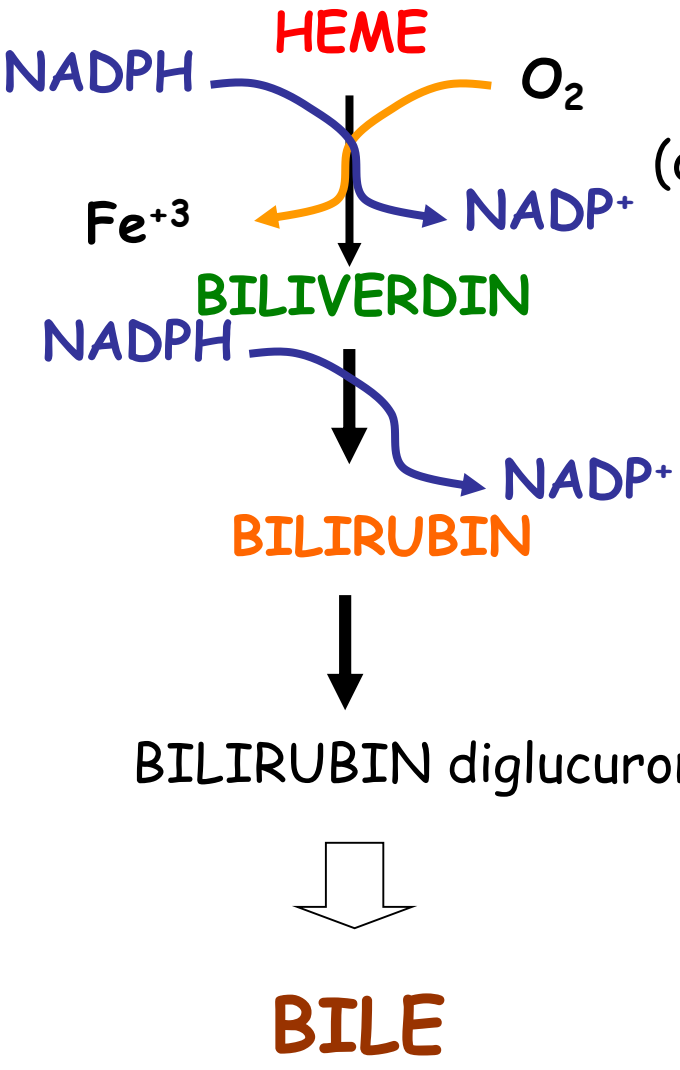
Function of Haemoglobin



When there is a high concentration of oxygen e.g in the alveoli haemoglobin combines with oxygen to form oxyhaemoglobin. When the blood reaches the tissue which have a low concentration of oxygen the haemoglobin dissociates with the oxygen and the oxygen is released into body tissues

Heme Degradation

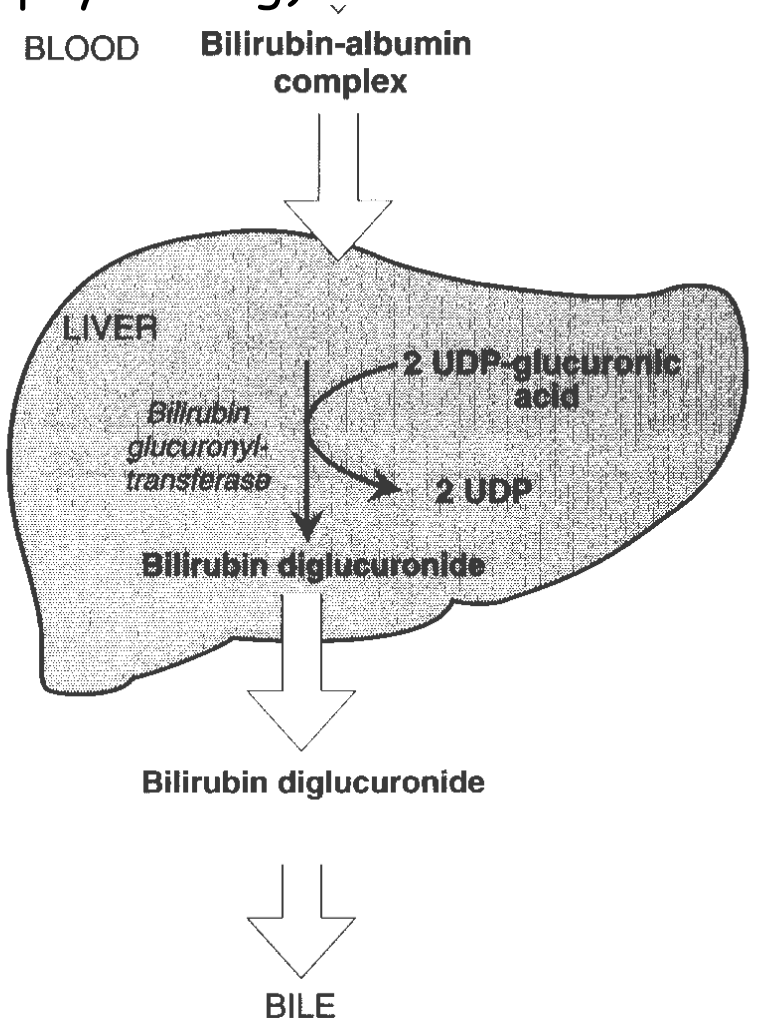
Heme Catabolism



BILIRUBIN diglucuronide

BILE

(opens the porphyrin ring)



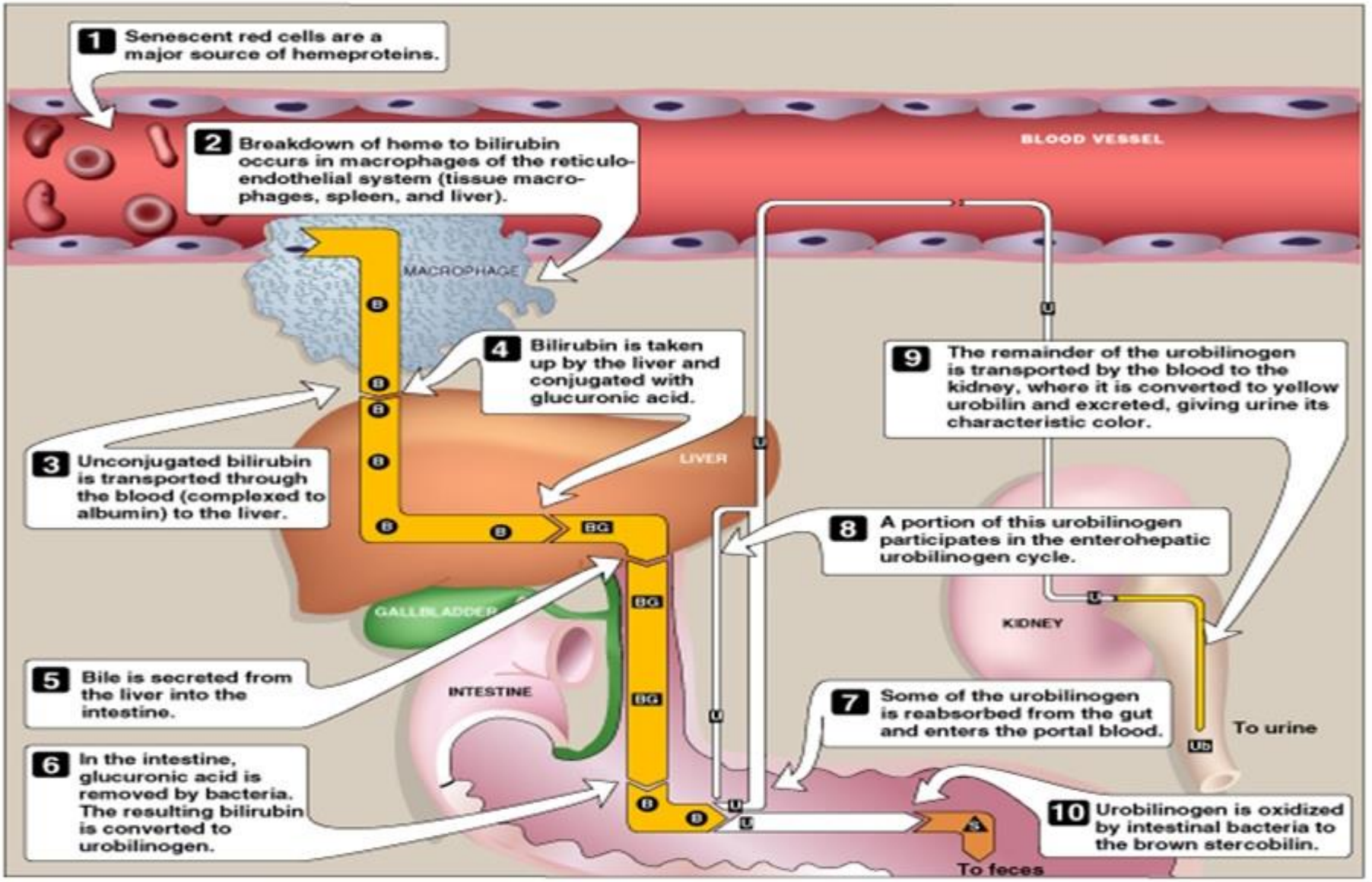
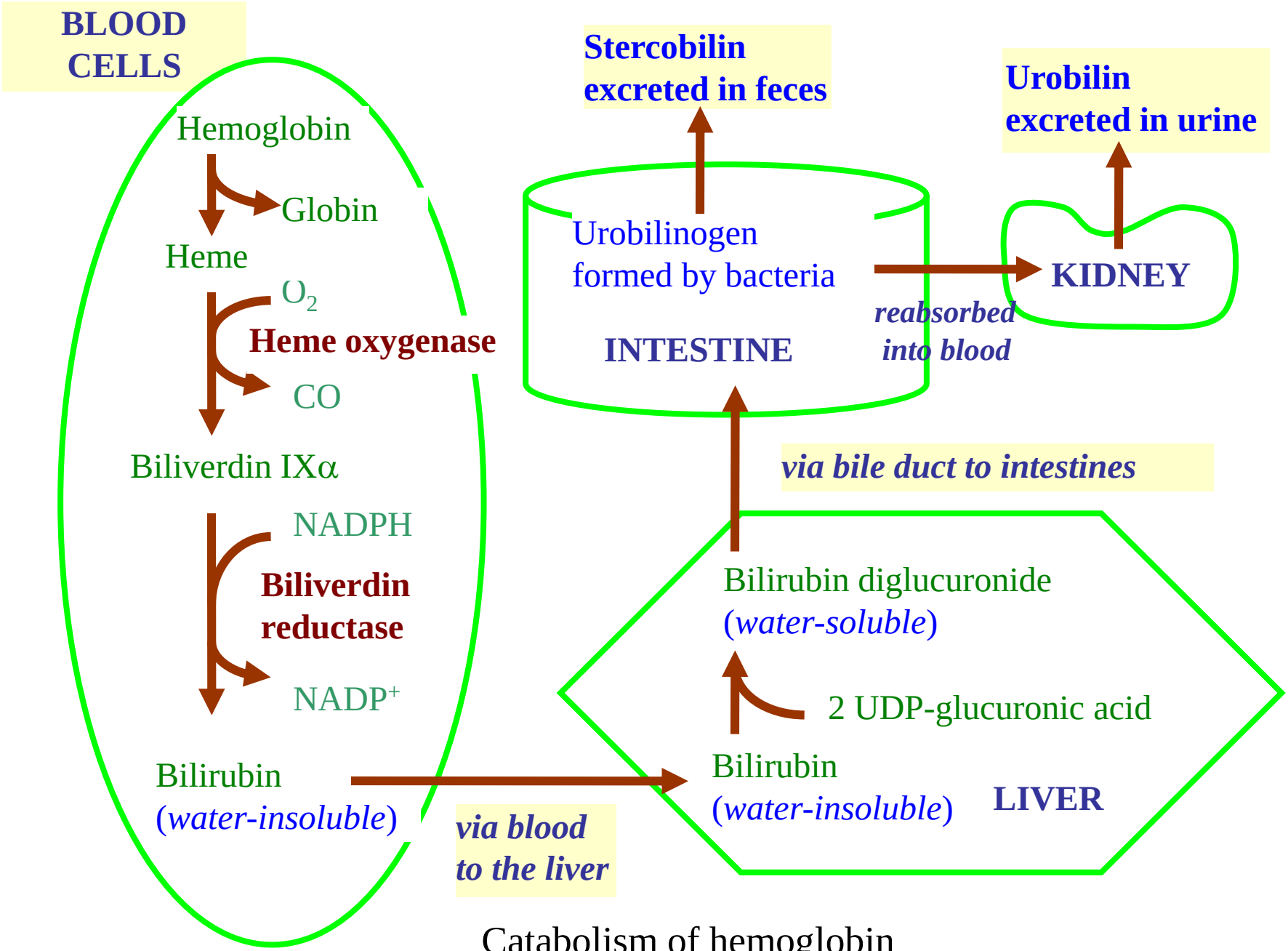


Figure 21.9
Catabolism of heme B = bilirubin; BG = bilirubin diglucuronide; U = urobilinogen; Ub = urobilin; S = stercobilin.



Catabolism of hemoglobin

Jaundice (icterus)

hyperbilirubinemia

- causes yellow color of skin, nail beds and sclerae

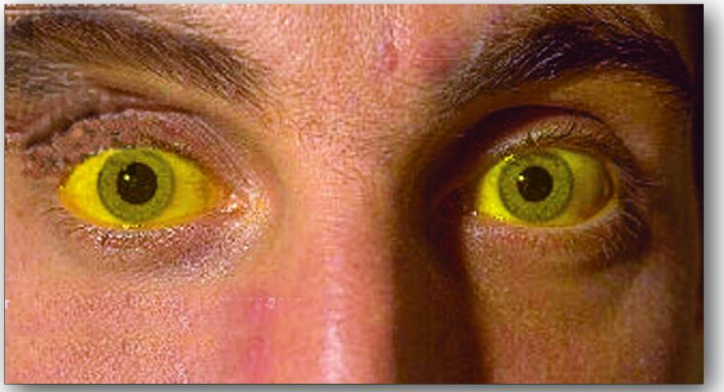
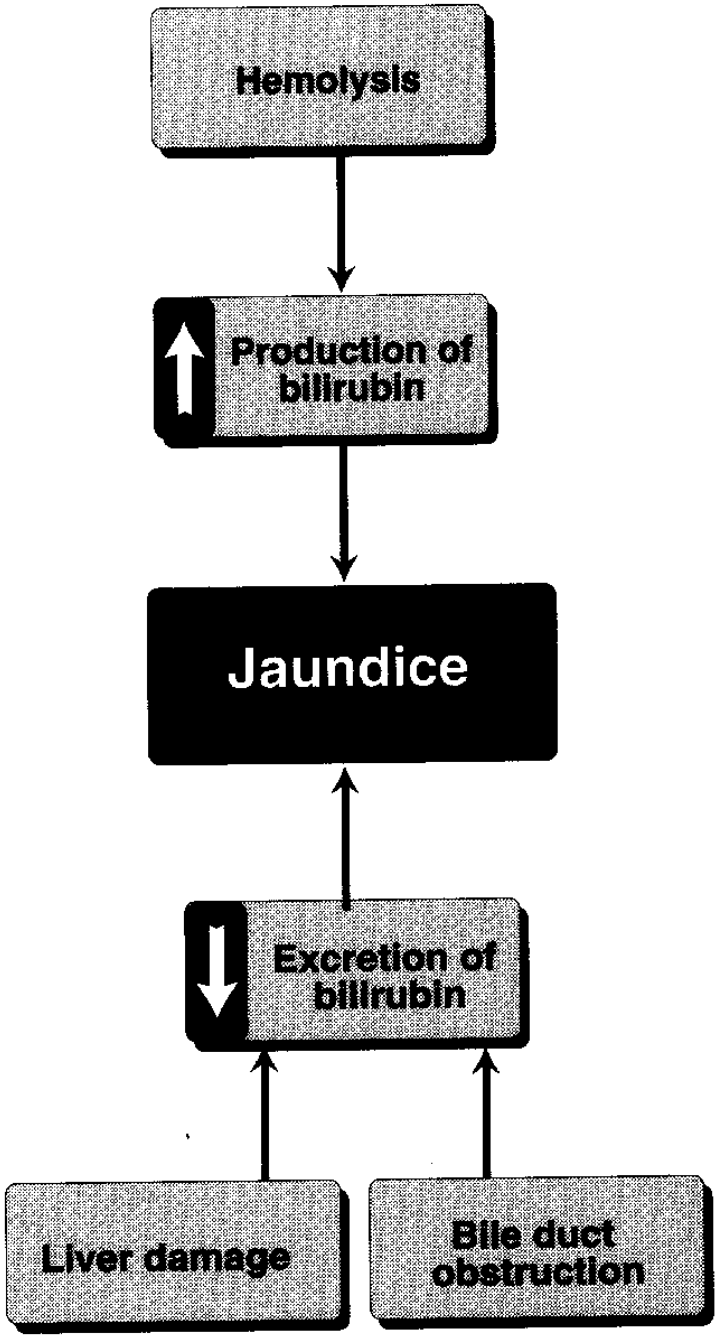


Figure 21.10
Jaundiced patient, with the sclerae of his eyes appearing yellow.

- not a disease, but symptom of underlying disorders



Jaundice

Hyperbilirubinemia:

Two forms:

Direct bilirubin: Conjugated with glucoronic acid

Indirect bilirubin: unconjugated, insoluble in water.

Types of Jaundice

hemolytic jaundice

- liver can handle 3000 mg bilirubin/day - normal is 300
- massive hemolysis causes more than can be processed
  - can’t be conjugated
  - increased bilirubin excreted into bile, urobilinogen is increased in blood, urine
  - unconjugated bilirubin in blood increases = jaundice

obstructive jaundice

- obstruction of the bile duct
  - tumor or bile stones
  - gastrointestinal pain - nausea
  - pale, clay-colored stools
  - can lead to liver damage and increased unconjugated bilirubin

Types of Jaundice

Hepatocellular Jaundice

- liver damage (cirrhosis or hepatitis) cause increased bilirubin levels in blood due to decreased conjugation
- conjugated bilirubin not efficiently exported to bile so diffuses into blood
- increased urobilinogen in enterohepatic circulation
    - so urine is darker and stool is pale, clay-colored
- Serum Aminotransferases (AST and ALT levels) are elevated
- nausea and anorexia

Van den Bergh reaction

1-This is a reaction between **bilirubin and Ehrlich diazo reagent** giving a **reddish purple** compound.

2- **Conjugated bilirubin** reacts directly with the reagent. Thus it is called: **direct bilirubin**

3- **Unconjugated bilirubin** does not react with the reagent directly except after addition of methyl alcohol. Thus it may be called: **indirect bilirubin**

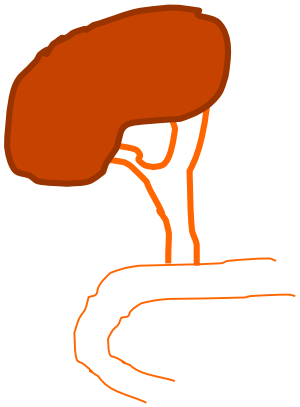
Serum Bilirubin – Applications

Type of Bilirubin	van den Bergh Reaction	Class of Jaundice	Causes
Unconjugated	Indirect positive	Pre-hepatic or Hemolytic	Neonatal jaundice Sickle cell anaemia Thalassemia Gilbert's syndrome Crigler-Najjar syndrome Mismatch blood transfusion
Both Unconjugated and Conjugated	Biphasic	Hepatic	Viral hepatitis Alcoholic hepatitis Drug induced hepatitis
Conjugated	Direct positive	Post-hepatic or Obstructive	Gall stones Biliary atresia Tumours of biliary tract Carcinoma head of pancreas

Examples of hyperbilirubinemia

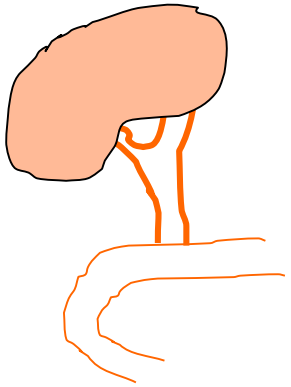
A. Hemolytic anemia

excess hemolysis



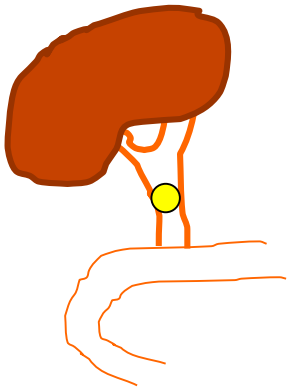
- ↑ unconjugated bilirubin (in blood)
- ↑ conjugated bilirubin (released to bile duct)

B. Hepatitis



- ↑ unconjugated bilirubin (in blood)
- ↑ conjugated bilirubin (in blood)

C. Biliary duct stone



- ↑ unconjugated bilirubin (in blood)
- ↑ conjugated bilirubin (in blood)

Table 2: Differences between the Three Basic Types of Jaundice

Features	Haemolytic Jaundice	Hepatocellular Jaundice	Extrahepatic Cholestatic Jaundice
Colour of urine	Pale	High coloured	High coloured
Colour of stools	Normal	Normal to dark	Pale coloured
Pruritus	-	-/+	++
Jaundice	Lemon yellow	Deep yellow	Orange yellow/greenish yellow
Antecedent history	H/o anaemia requiring blood transfusions (BT)	Injection/BT/tattoos contact with a jaundiced patient	Biliary surgery or gall stones
Family history	Anaemia	Jaundice	Gall stones
Urine urobilinogen	++	+	-
Haemogram	Evidence of haemolysis	Normal	Normal
Serum bilirubin	↑ <5 mg/dL (usually) (unconjugated)	↑ - ↑↑↑ (mixed)	↑ - ↑↑↑ (predominantly conjugated)
Serum transaminases	Normal	↑ - ↑↑↑	Normal/↑
Alkaline phosphatase	Normal	Normal/↑	↑↑
Ultrasonogram/CT	Hepatosplenomegaly liver ↑/normal spleen ↑ - ↑↑↑	Liver ↑/N spleen N/↑	Liver ↑↑ with dilated ducts

Jaundice in Newborns

premature babies often accumulate bilirubin due to late onset of expression of bilirubin glucuronyltransferase

- maximum expression (adult level) at ~ 4 weeks
- excess bilirubin can cause toxic encephalopathy (kernicterus)
- treated with blue fluorescent light
    - converts bilirubin to more polar compound
        - can be excreted in bile without conjugation
- Crigler - Najjar syndrome is deficiency in bilirubin glucuronyltransferase

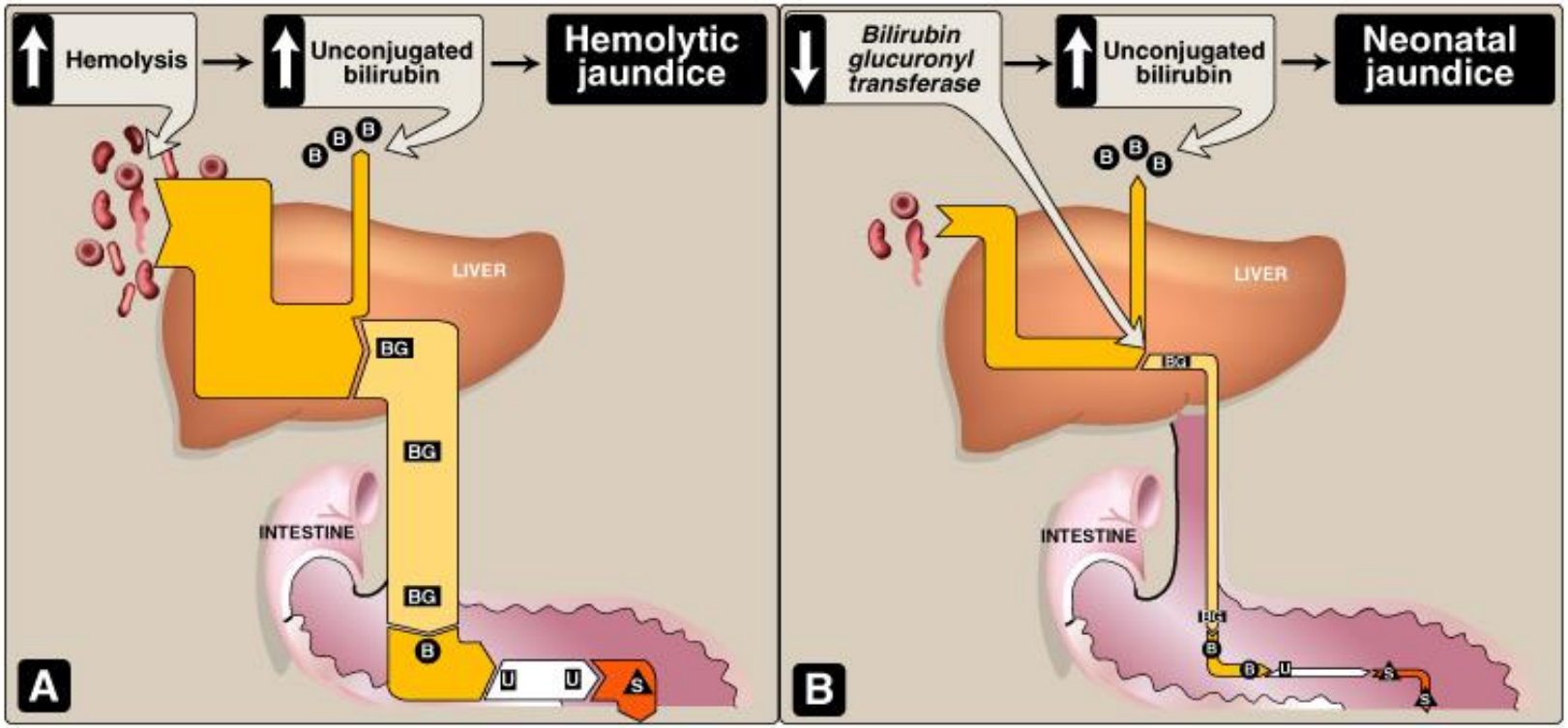


Figure 21.11

Alterations in the metabolism of heme. A. Hemolytic jaundice. B. Neonatal jaundice. [Note: The enterohepatic circulation of urobilinogen is omitted for simplicity.] BG = bilirubin glucuronide; B = bilirubin; U = urobilinogen; S = stercobilin.

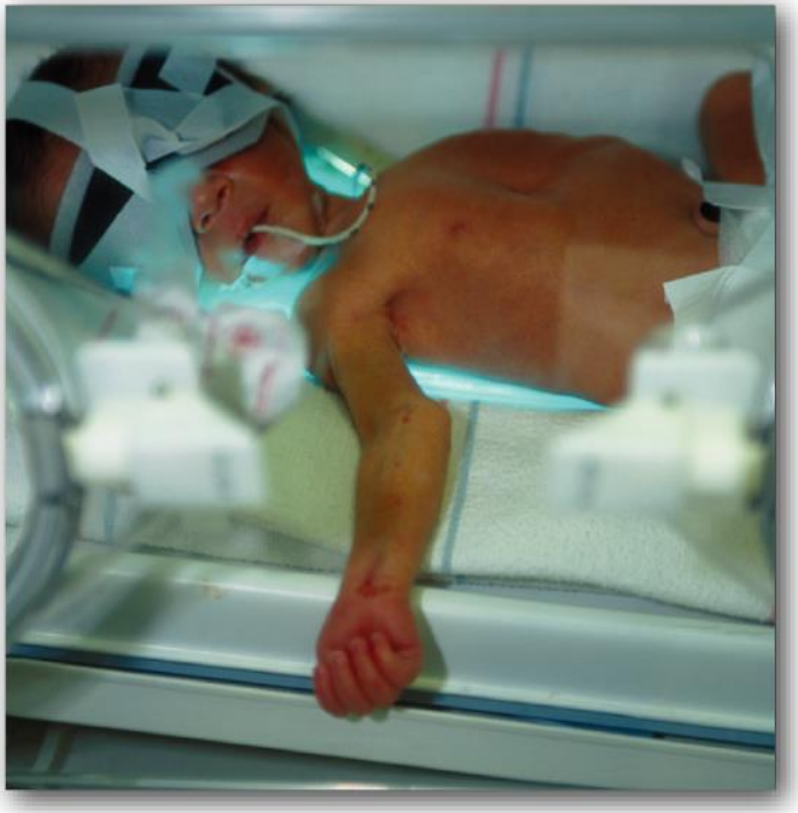
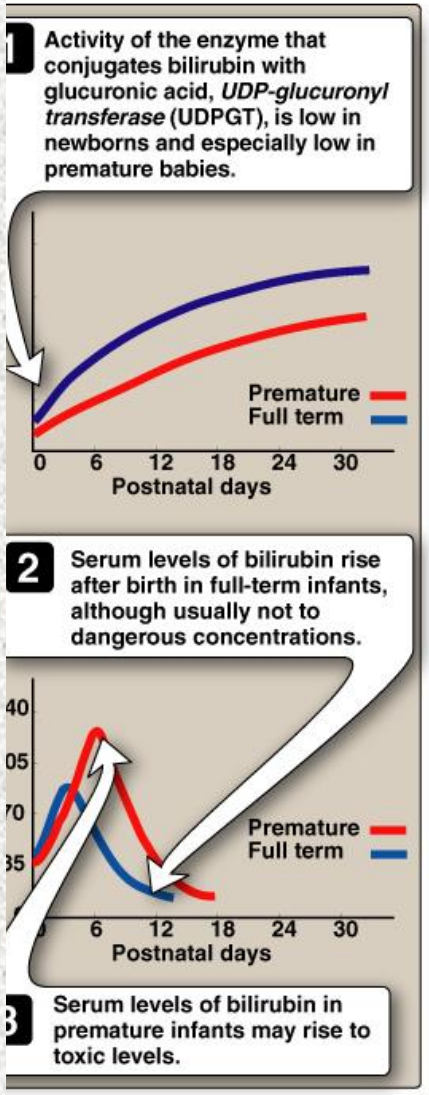


Figure 21.13
Phototherapy in neonatal jaundice.



ure 21.12
natal jaundice.