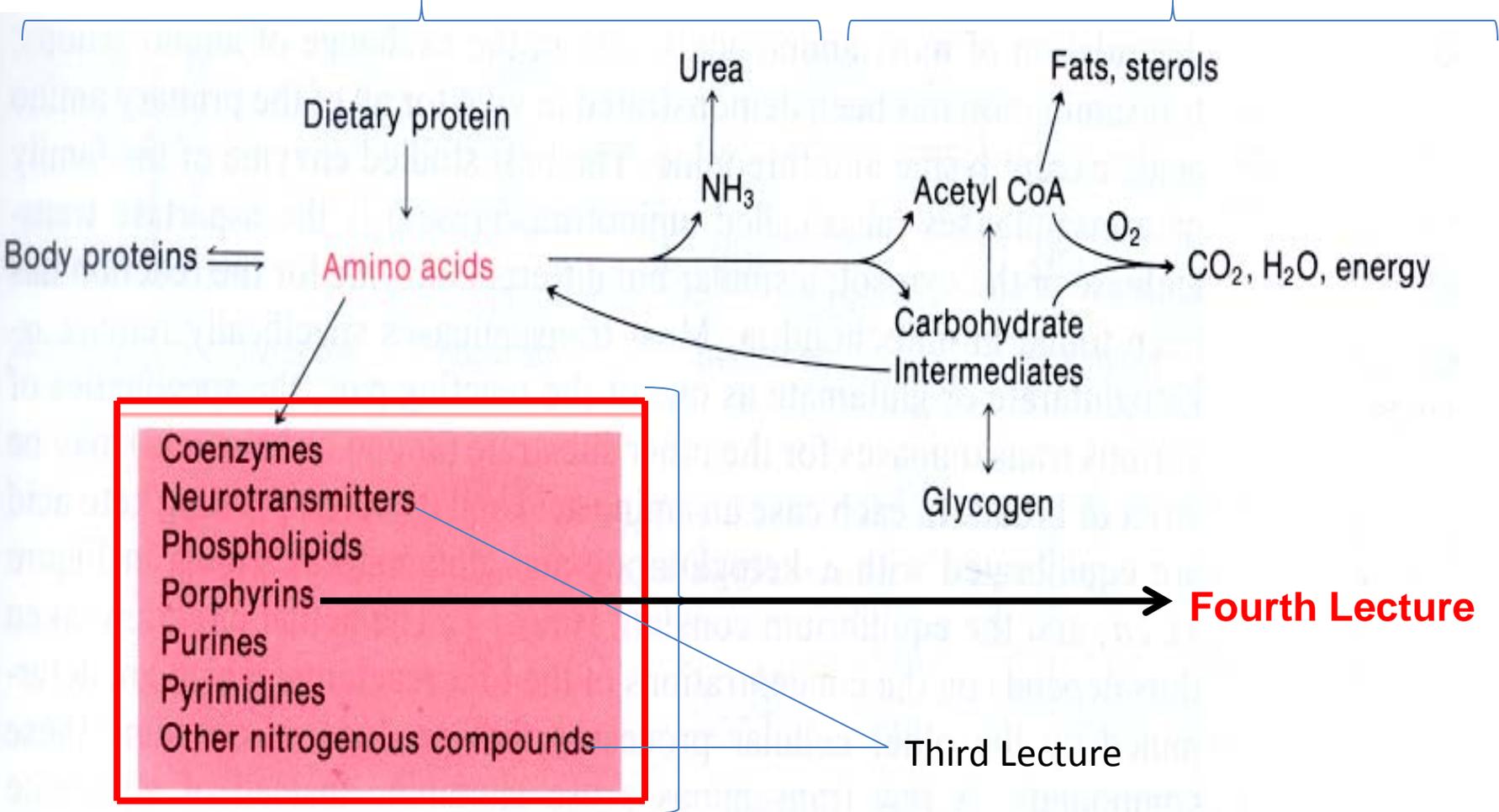


Conversion of Amino Acids to Specialized Products

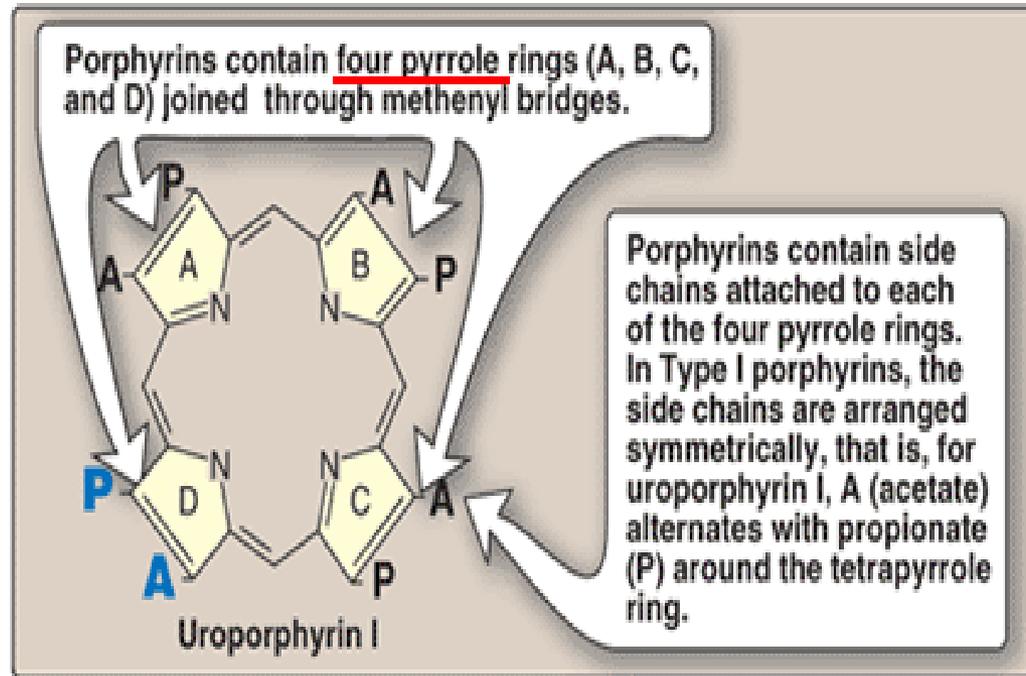
First Lecture

Second lecture



Structure of porphyrins

Porphyrins are cyclic compounds that readily bind metal ions (metalloporphyrins)—usually Fe^{2+} or Fe^{3+} .



Porphyrins vary in the nature of the side chains that are attached to each of the four pyrrole rings.

Uroporphyrin contains acetate ($-\text{CH}_2-\text{COO}^-$) and propionate ($-\text{CH}_2-\text{CH}_2-\text{COO}^-$) side chains;

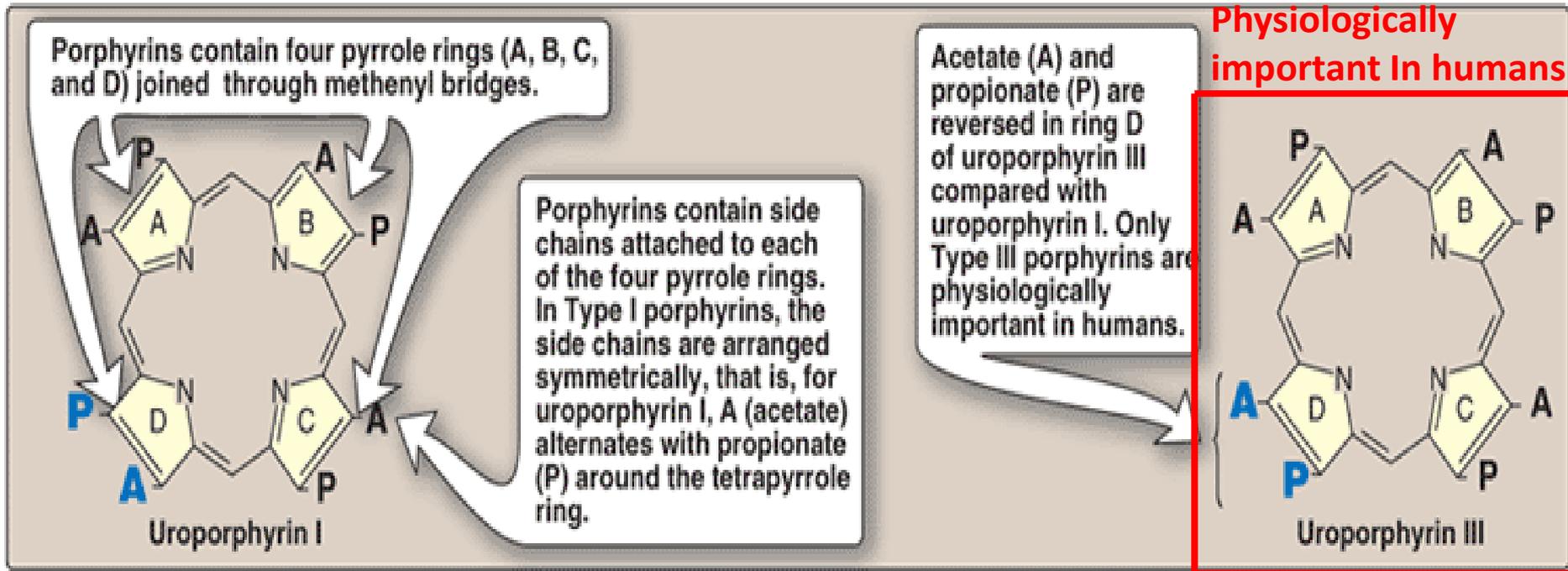
Coproporphyrin contains methyl ($-\text{CH}_3$) and propionate groups;

Protoporphyrin IX (and heme) contains vinyl ($-\text{CH}=\text{CH}_2$), methyl, and propionate groups

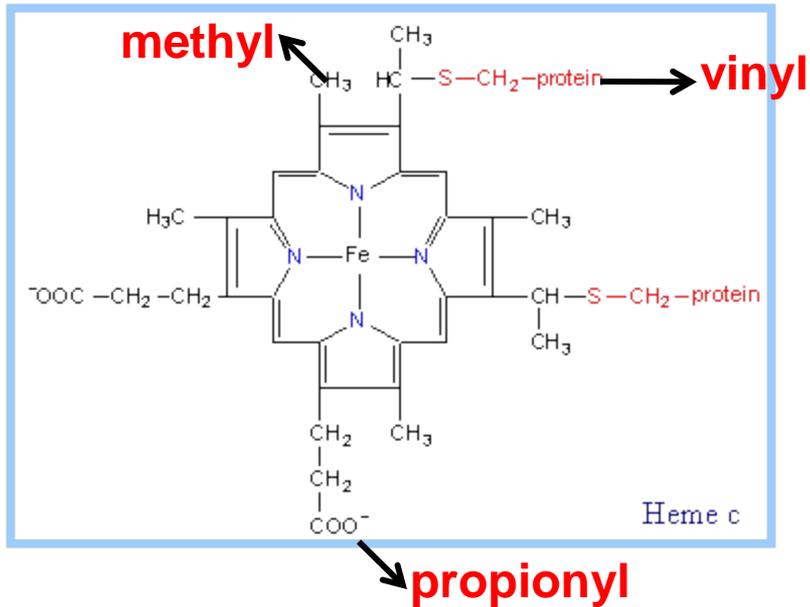
Structure of Porphyrins

Distribution of side chains: The side chains of porphyrins can be ordered around the tetrapyrrole nucleus in four different ways, designated by Roman numerals I to IV.

Only Type III porphyrins, which contain an asymmetric substitution on ring D are physiologically important in humans.



Heme



- 1) Four Pyrrole rings linked together with methenyle bridges;
- 2) Three types of side chains are attached to the rings; arrangement of these side chains determines the activity;
- 3) Asymmetric molecule
- 3) Porphyrins bind metal ions to form metalloporphyrins.

Heme is a prosthetic group for hemoglobin, myoglobin, the cytochromes, catalase and tryptophan pyrrolase

Biosynthesis of Heme

The major sites of heme biosynthesis are the **liver**, which synthesizes a number of heme proteins (particularly cytochrome P450), and the **erythrocyte-producing cells of the bone marrow**, which are active in hemoglobin synthesis.

Heme synthesis occurs in all cells due to the requirement for heme as a prosthetic group on enzymes and electron transport chain.

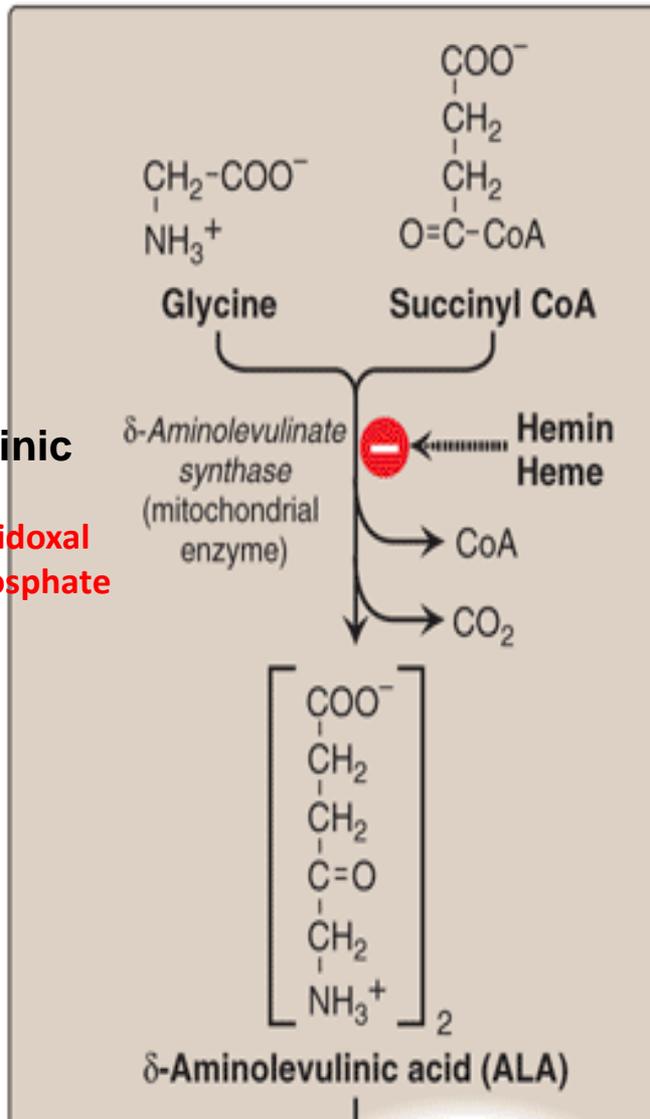
The initial reaction and the last three steps in the formation of porphyrins occur in **mitochondria**, whereas the intermediate steps of the biosynthetic pathway occur in the **cytosol**.

[Note: Mature red blood cells lack mitochondria and are unable to synthesize heme.]

Biosynthesis of Heme

Step I: Formation of δ -aminolevulinic acid (ALA)

δ -aminolevulinic acid (ALA) synthase
Pyridoxal Phosphate



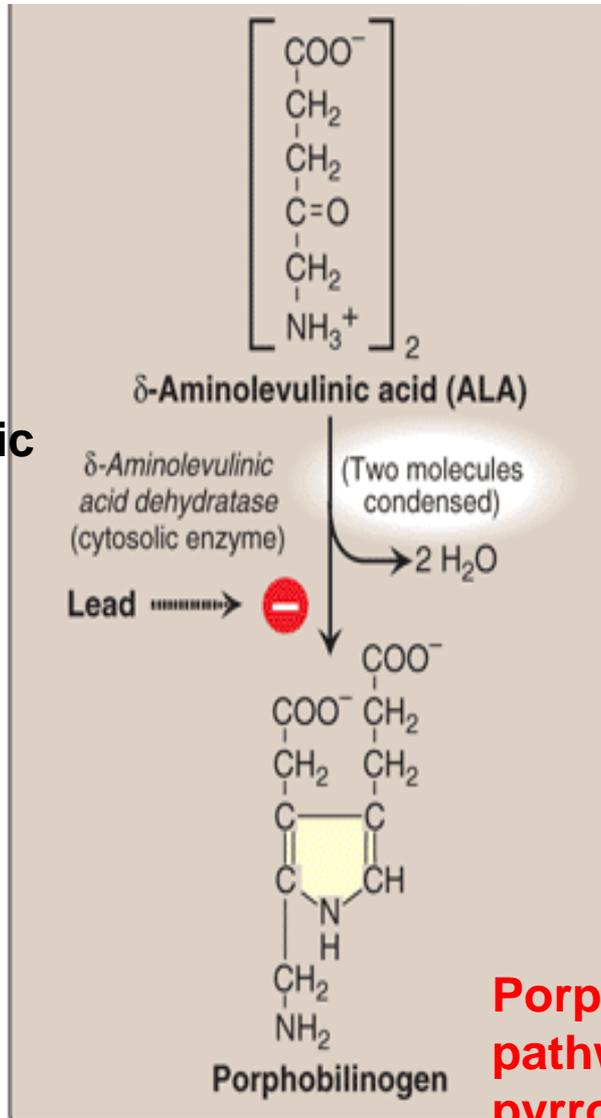
ALA synthase activity (transcription) is modulated by

- Heme (inhibits ALA synthase)
- Drugs (e.g. antifungals and Anticonvulsants) (enhances the ALA activity)

↓
Cytochrome P450 monooxygenase

Biosynthesis of Heme

Step II: Formation of porphobilinogen:



δ -aminolevulinic acid (ALA) dehydratase

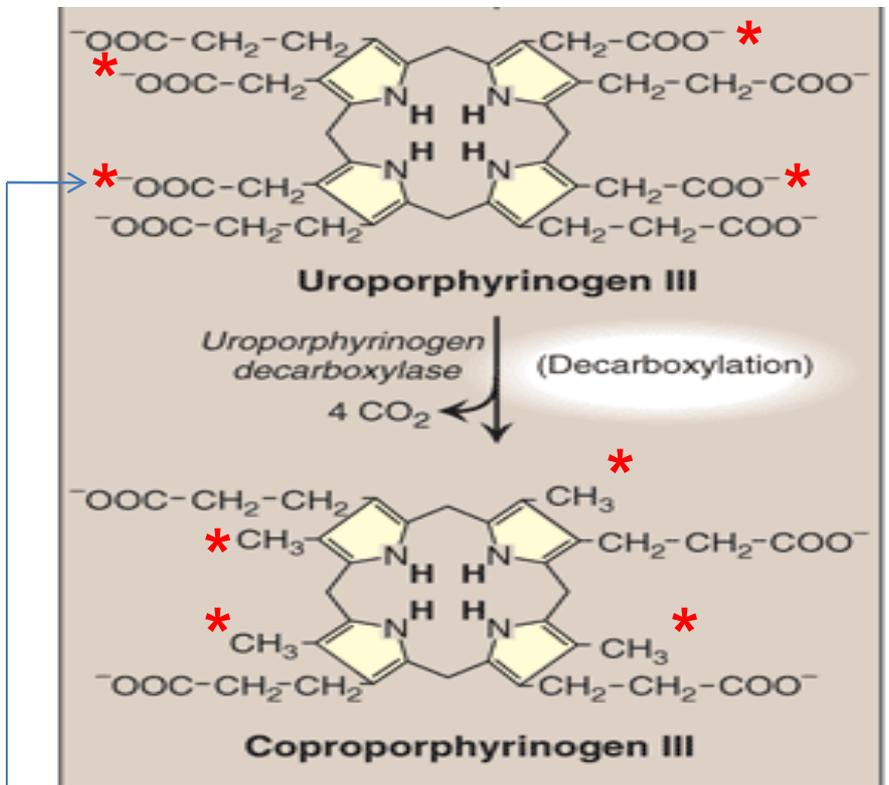
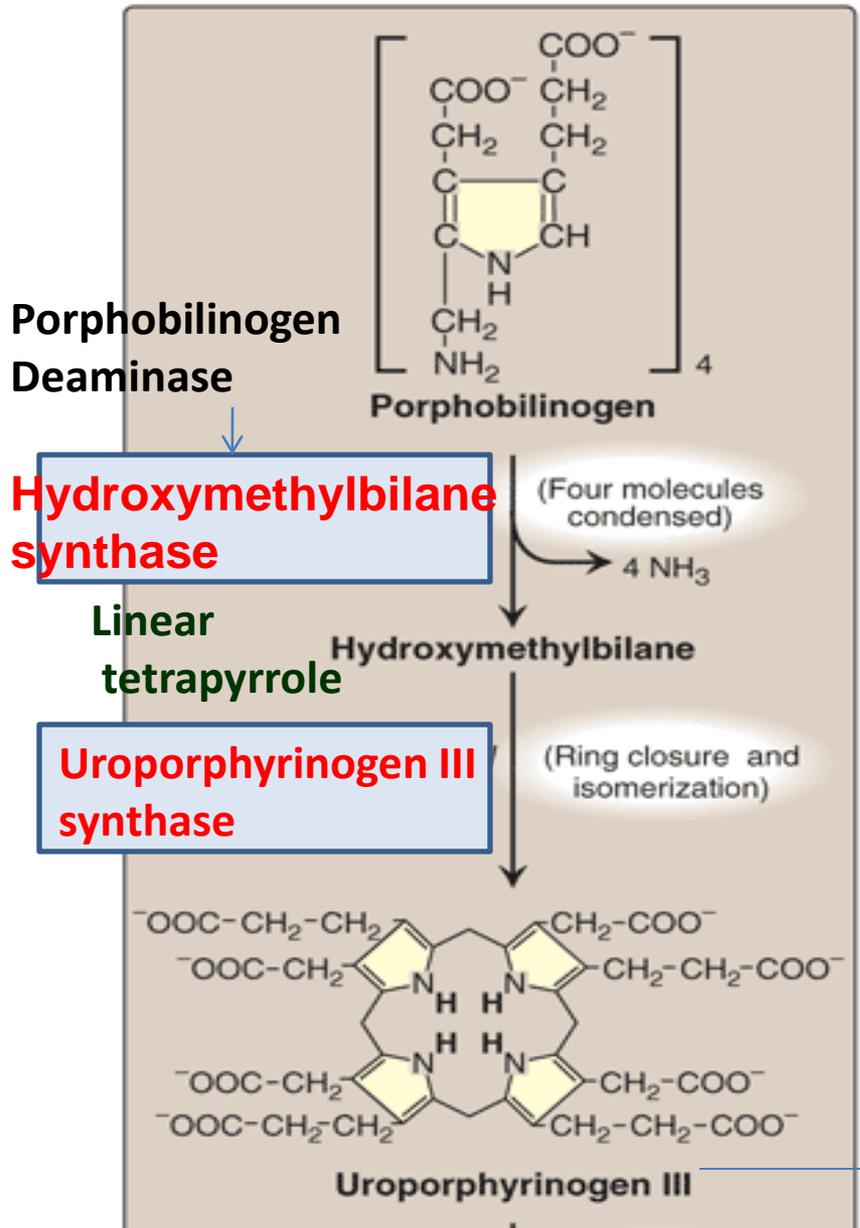
The condensation of two molecules of ALA to form porphobilinogen by ALA dehydratase.

Reaction is extremely sensitive to inhibition by **heavy metal ions**. This inhibition is, in part, responsible for the elevation in ALA and the anemia seen in lead poisoning.

Porphobilinogen (PBG) is the first pathway intermediate that includes a pyrrole ring.

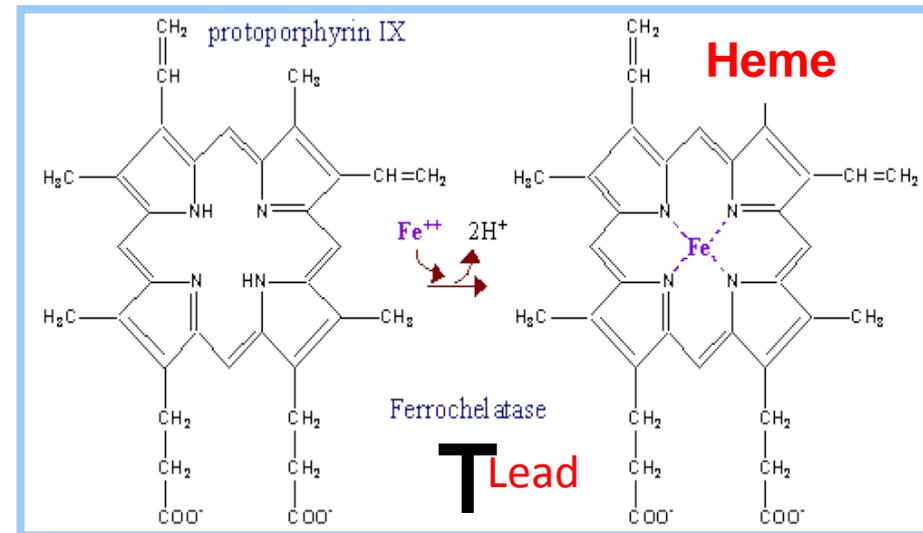
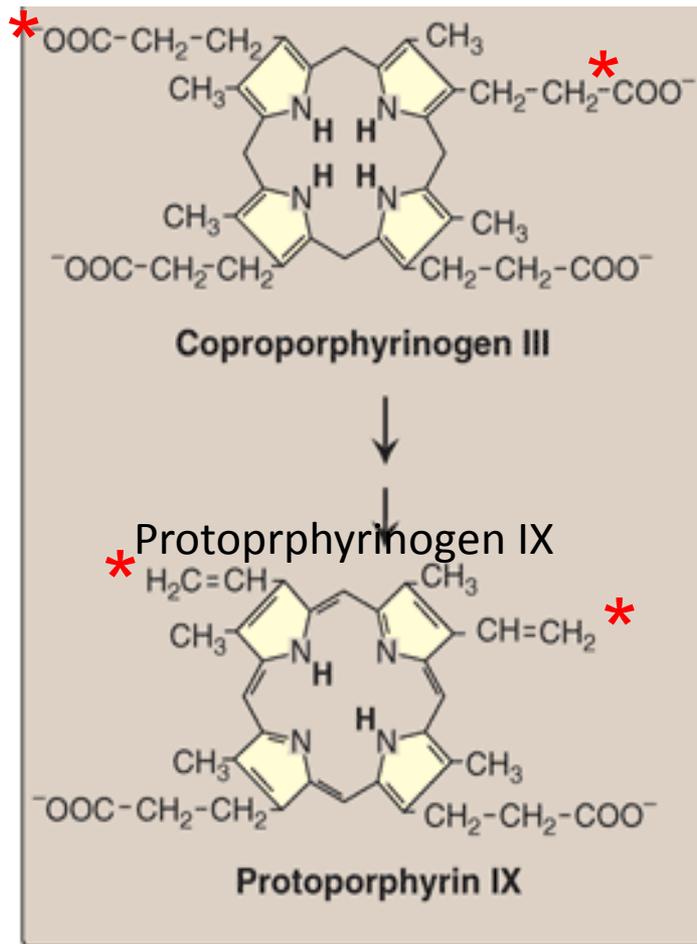
Biosynthesis of Heme

Step III : Formation of uroporphyrinogen



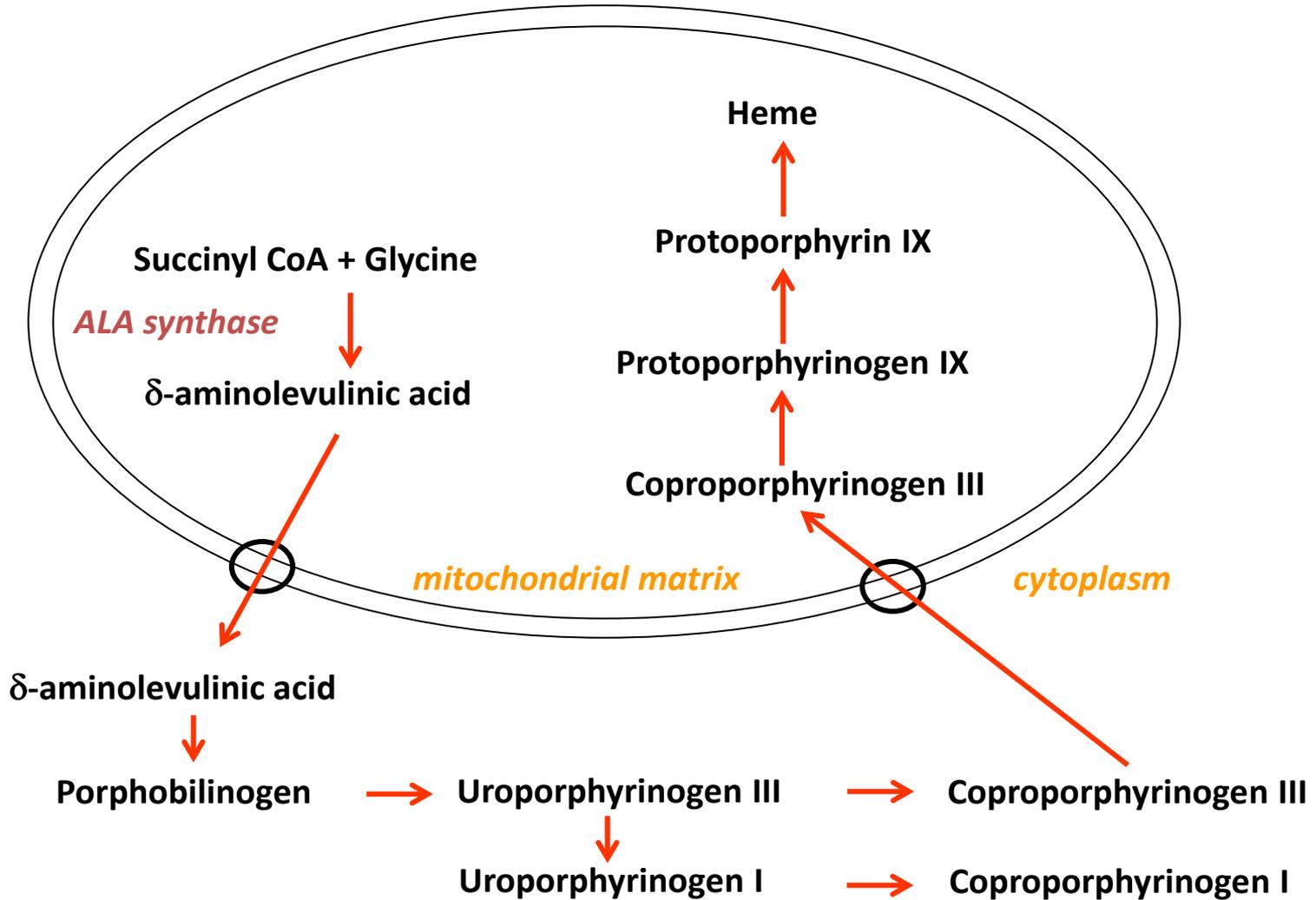
Biosynthesis of heme

Step IV Formation of heme



Coproporphyrinogen III enters the mitochondrion, and two propionate side chains are decarboxylated to vinyl groups generating protoporphyrinogen IX, which is oxidized to protoporphyrin IX.

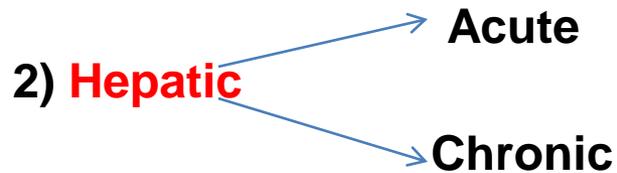
Overview of Heme Synthesis



Porphyrias

Porphyrias are rare, inherited (or occasionally acquired) defects in heme synthesis, resulting in the accumulation and increased excretion of porphyrins or porphyrin precursors (purple color pigment in urine).

1) Erythropoietic Porphyria



Hepatic Porphyria (Acute)

Acute intermittent porphyria (AIP)- Uroporphyrinogen synthase deficiency with high ALA or PBG in urine and serum.

Variagate porphyria (VP)- Protoporphyrinogen oxidase deficiency with high fecal levels of protoporphyrin and coproporphyrin.

Hereditary coproporphyrria - Coproporphyrinogen oxidase deficiency with high urinary and/or fecal levels of coproporphyrins.

ALA dehydratase deficient porphyria (rare).

Erythropoietic protoporphyria (EPP)- Ferrochelatase deficiency.

Symptoms: Acute attacks of gastrointestinal, neurologic/psychiatric, and cardiovascular symptoms.

Porphyria cutanea tarda

Chronic porphyria

- It is a chronic disease of the liver and erythroid tissues.
- The disease is associated with a deficiency in **uroporphyrinogen decarboxylase**.
- Clinical expression of the enzyme deficiency is influenced by various factors, such as hepatic iron overload, exposure to sunlight, and the presence of hepatitis B or C, or HIV infections.
- Clinical onset is typically during the fourth or fifth decade of life.
- Porphyrin accumulation leads to cutaneous symptoms and urine that is red to brown in natural light and pink to red in fluorescent light.

Skin lesions



Normal



Erythropoietic porphyrias

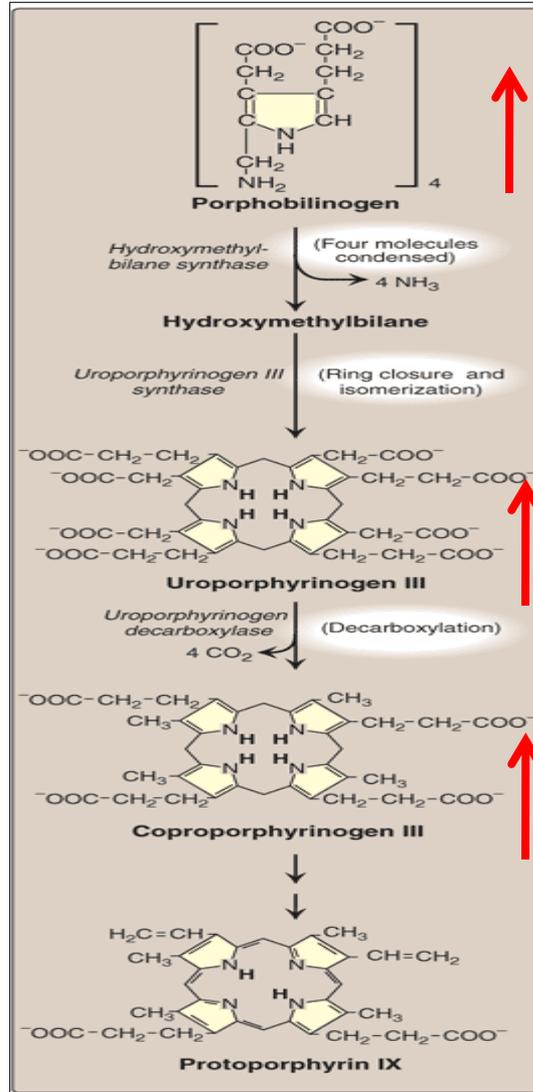
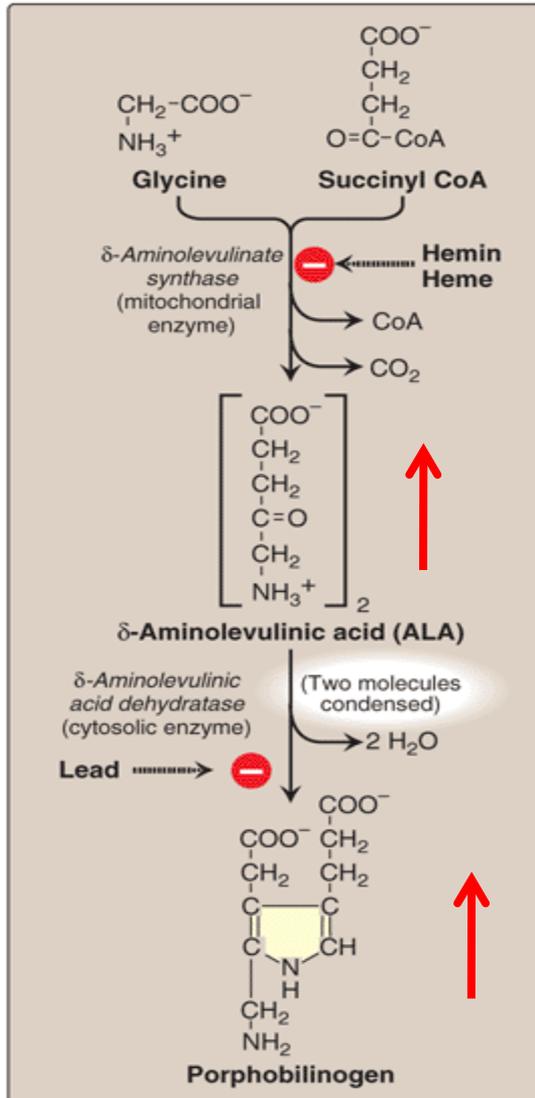
Congenital erythropoietic porphyria and erythropoietic protoporphyria)

They are characterized by skin rashes and blisters that appear in early childhood.

The diseases are complicated by cholestatic liver cirrhosis and progressive hepatic failure.

Increased ALA synthase activity is the culprit

Is caused due to the decreased Heme synthesis



Leads to increased synthesis of intermediates that occur prior to the genetic block. The accumulation of these toxic intermediates is the major pathophysiology of the porphyrias.

Treatment

The severity of symptoms of the porphyrias can be diminished by intravenous injection of hemin, which decreases the synthesis of ALA synthase.

Avoidance of sunlight and ingestion of β -carotene (a free-radical scavenger) are also helpful.

Acquired Porphyrias

Lead poisoning

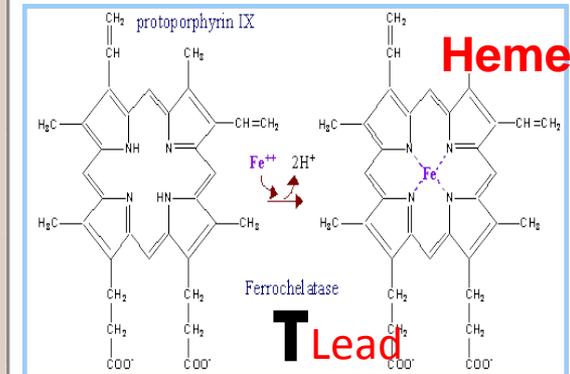
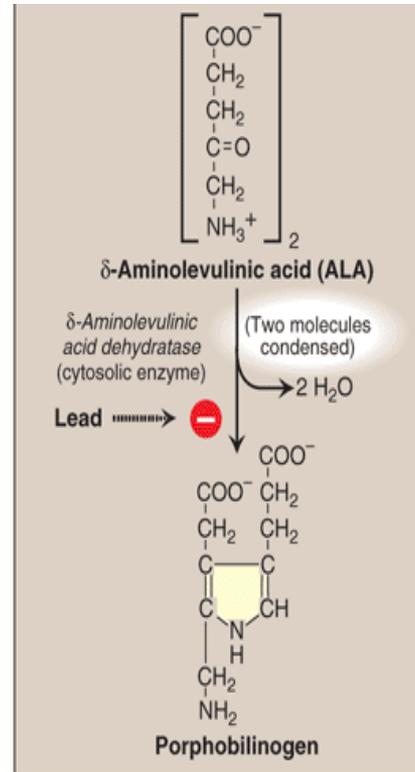
- inhibition of ferrochelatase and ALA dehydratase
- displaces Zn^{+2} at enzyme active site

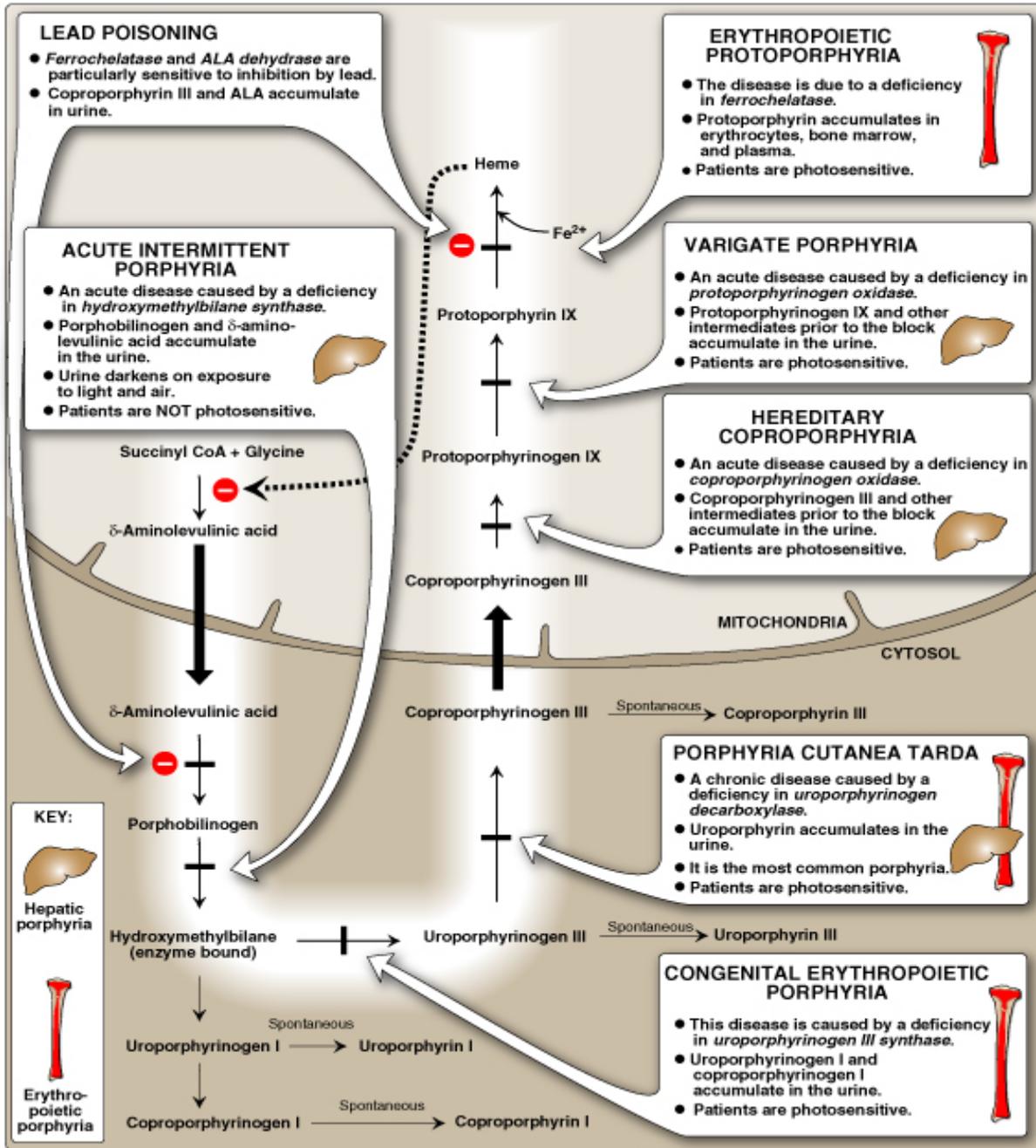
Children

- developmental defects
- drop in IQ
- hyperactivity
- insomnia
- many other health problems

Adults

- severe abdominal pain
- mental confusion
- many other symptoms





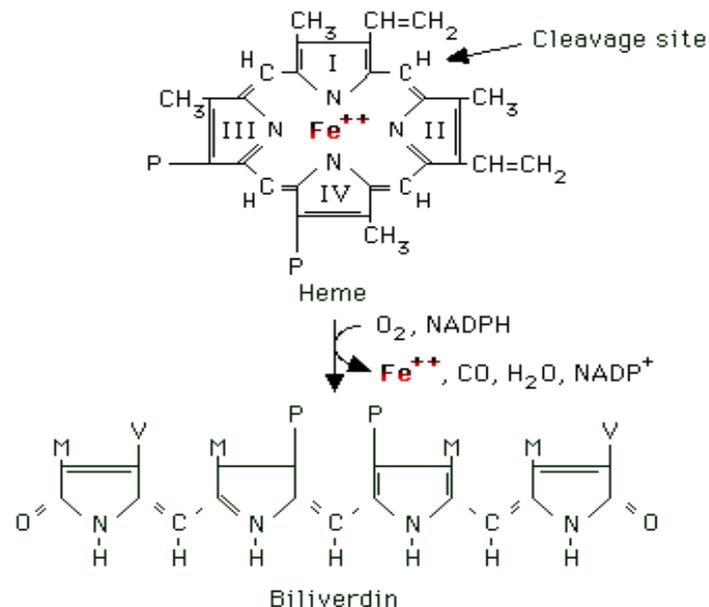
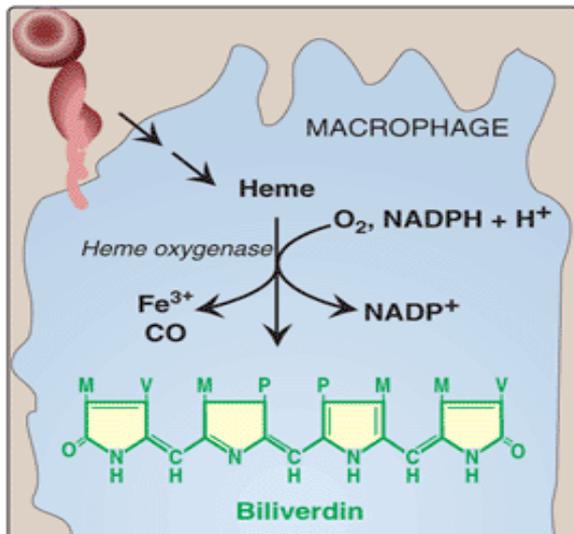
Degradation of heme

The first reaction is cleavage of the heme ring by a microsomal heme oxygenase.

This enzyme adds a hydroxyl group to the methenyl bridge between two pyrrole rings (I and II), with a concomitant oxidation of ferrous iron to Fe^{3+} .

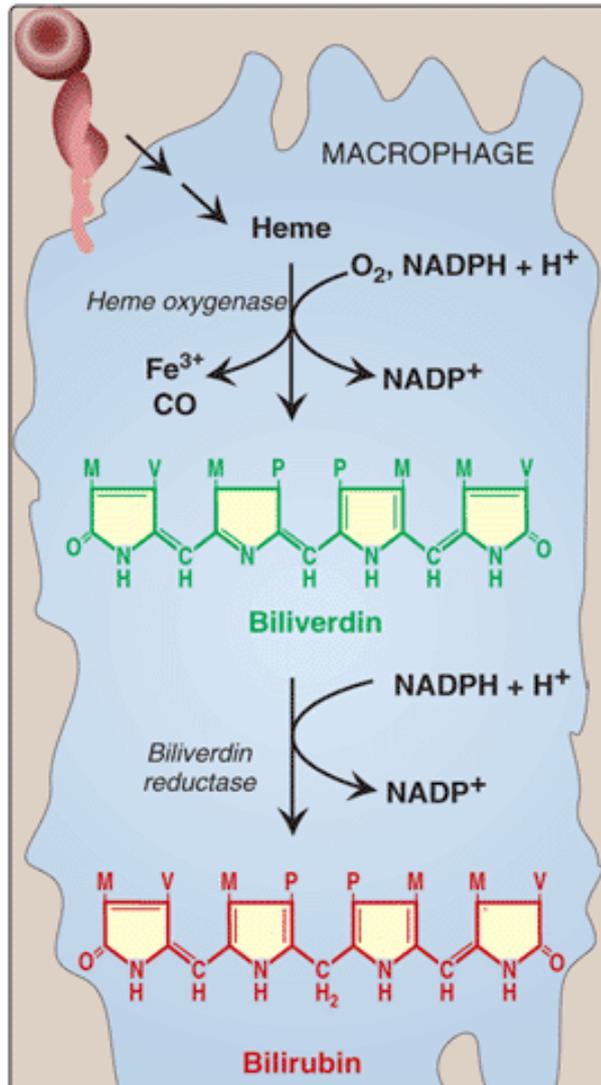
A second oxidation by the same enzyme system results in cleavage of the porphyrin ring. The green pigment biliverdin is produced as ferric iron and CO are released

Reaction Catalyzed by Heme Oxygenase



Degradation of Heme

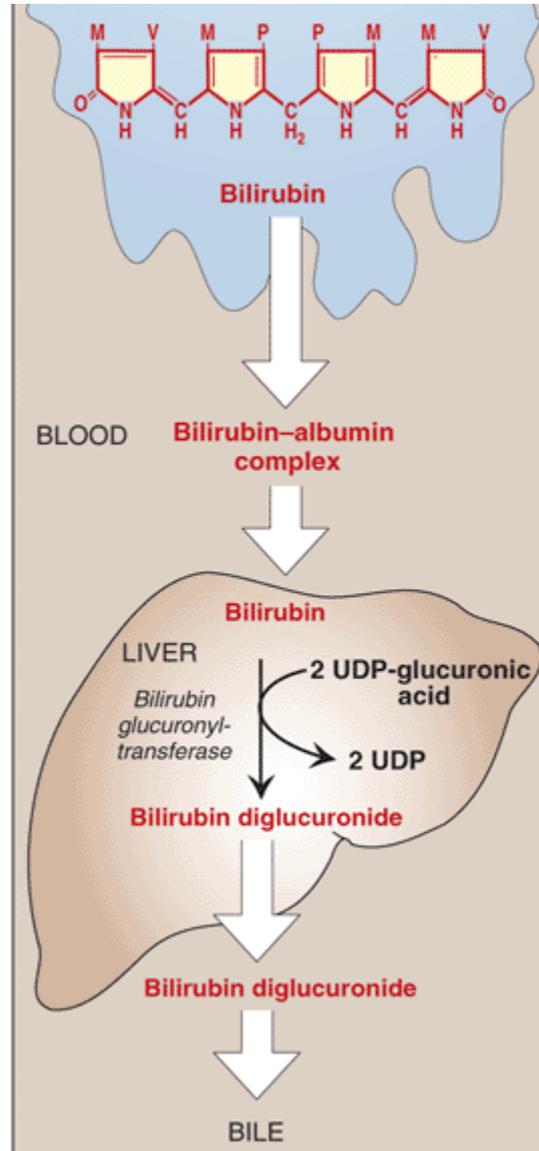
In the second reaction biliverdin reductase reduces the central methenyl bridge of biliverdin, producing bilirubin.



Biliverdin is reduced, forming the red-orange bilirubin. Bilirubin and its derivatives are collectively termed bile pigments.

[Note: The changing colors of a bruise reflect the varying pattern of intermediates that occur during heme degradation.]

Uptake of bilirubin by the liver

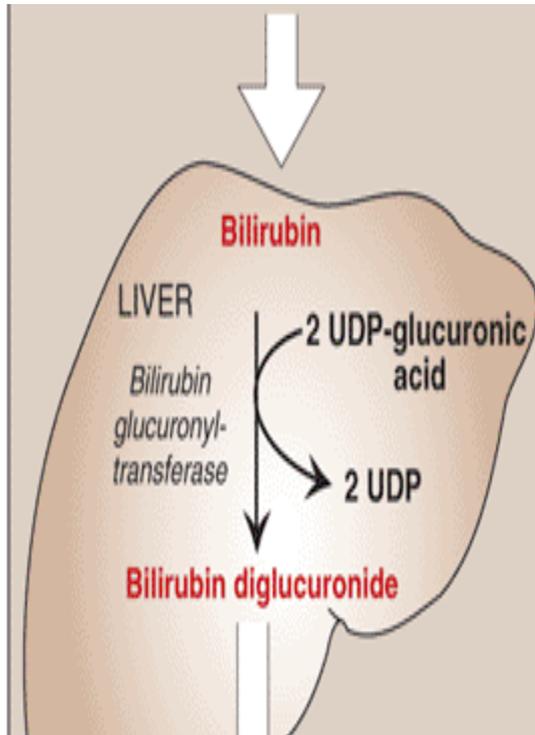


Bilirubin is only slightly soluble in plasma and, therefore, is transported to the liver by binding non-covalently to albumin.

[Note: Certain anionic drugs, such as salicylates and sulfonamides, can displace bilirubin from albumin, permitting bilirubin to enter the central nervous system. This causes the potential for neural damage in infants.]

Bilirubin dissociates from the carrier albumin molecule and enters a hepatocyte.

Formation of bilirubin diglucuronide

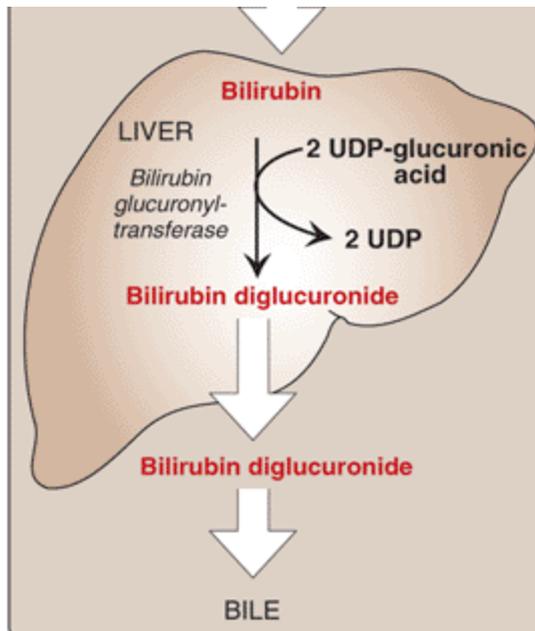


In the hepatocyte, the solubility of bilirubin is increased by the addition of two molecules of glucuronic acid.

The reaction is catalyzed by microsomal bilirubin glucuronyltransferase using uridine diphosphate-glucuronic acid as the glucuronate donor.

[Note: Varying degrees of deficiency of this enzyme result in Crigler-Najjar I and II and Gilbert syndrome, with Crigler-Najjar I being the most severe deficiency.]

Secretion of bilirubin into bile



Bilirubin diglucuronide (conjugated bilirubin) is actively transported into the bile.

Unconjugated bilirubin is normally not secreted.

This energy-dependent, rate-limiting step is susceptible to impairment in liver disease.

[Note: A deficiency in the protein required for transport of conjugated bilirubin out of the liver results in Dubin-Johnson syndrome.]

Formation of urobilins in the intestine

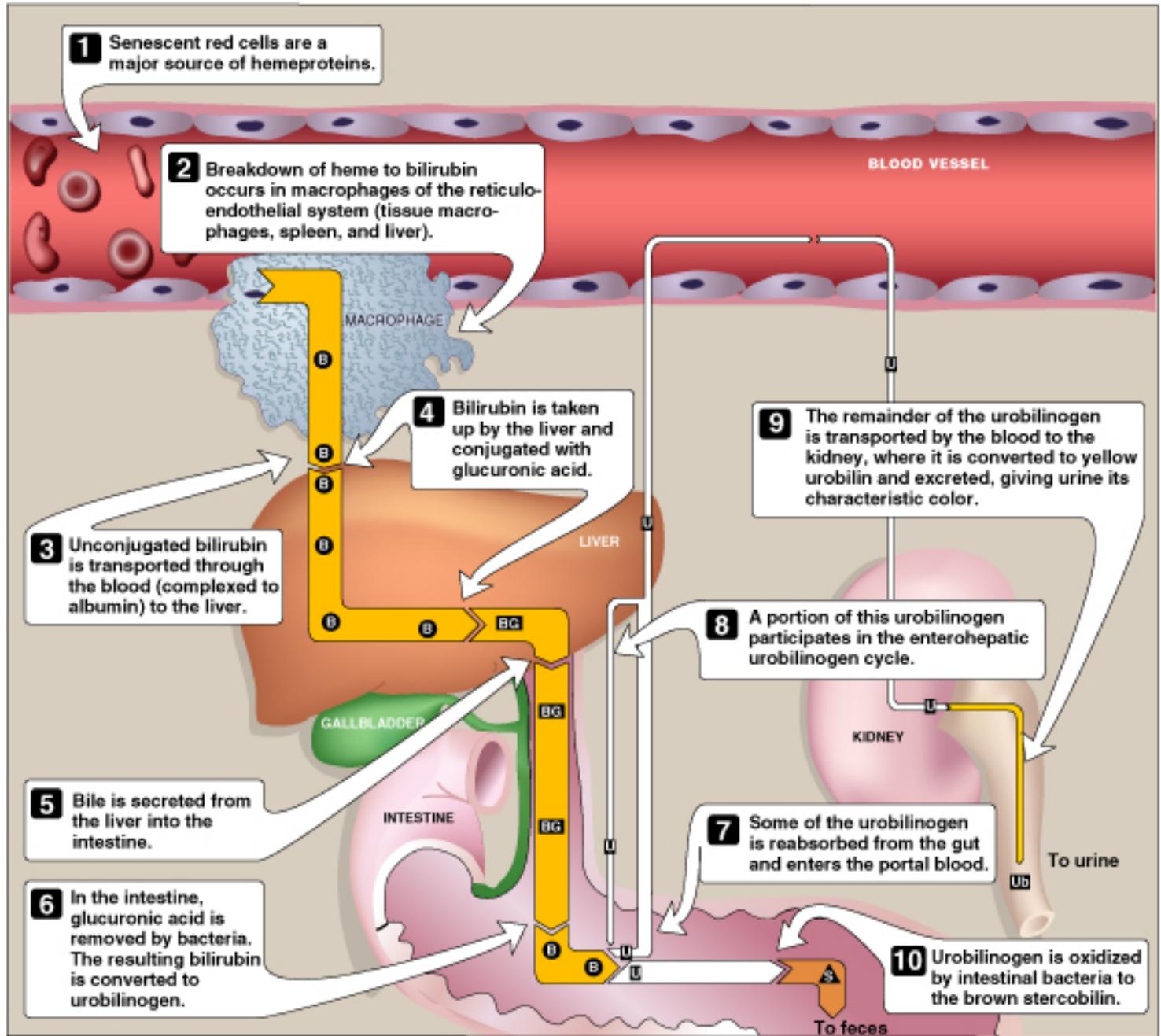
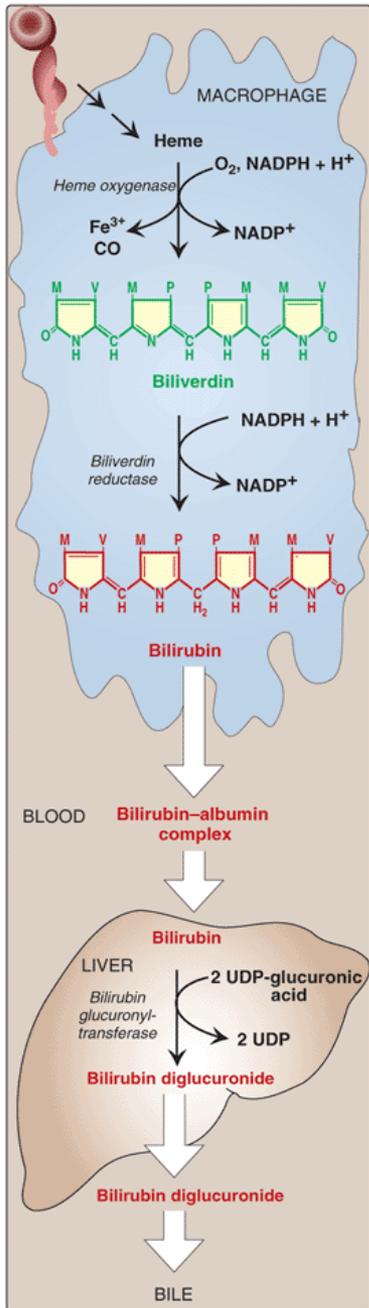
Bilirubin diglucuronide is hydrolyzed and reduced by bacteria in the gut to yield urobilinogen, a colorless compound.

Most of the urobilinogen is oxidized by intestinal bacteria to stercobilin, which gives feces the characteristic brown color.

However, some of the urobilinogen is reabsorbed from the gut and enters the portal blood. A portion of this urobilinogen participates in the enterohepatic urobilinogen cycle in which it is taken up by the liver, and then resecreted into the bile.

The remainder of the urobilinogen is transported by the blood to the kidney, where it is converted to yellow urobilin and excreted, giving urine its characteristic color.

Degradation of Heme



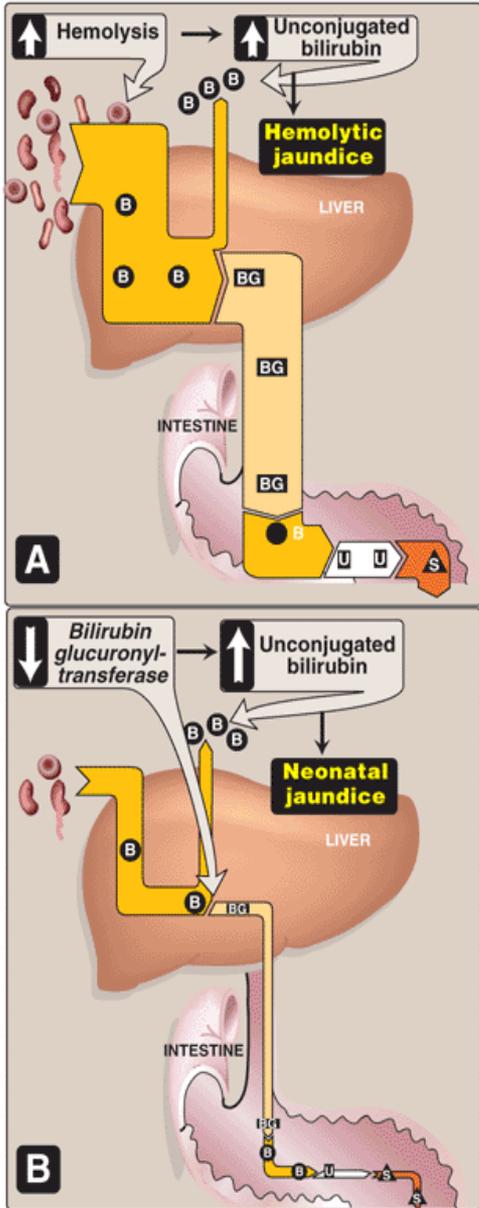
Jaundice

Defects in the degradation of Heme

Symptoms: Yellow color of skin, nail beds, and sclerae (whites of the eyes) caused by deposition of bilirubin, secondary to increased bilirubin levels in the blood



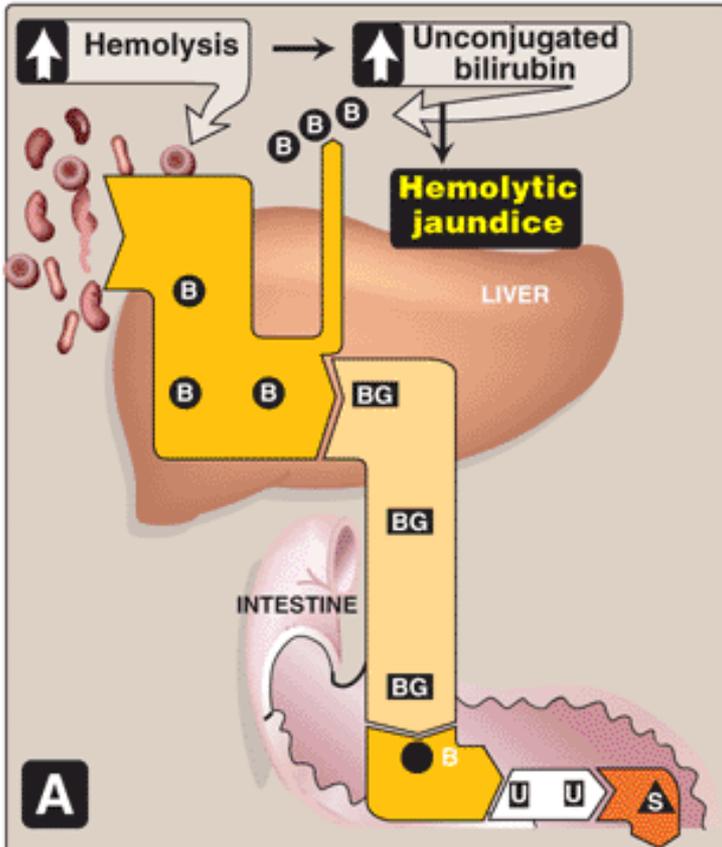
Types of Jaundice



Alterations in the metabolism of heme.

- A. Hemolytic jaundice.**
- B. Neonatal jaundice.**

Hemolytic Jaundice



Caused due to the Sickle cell anemia, pyruvate kinase or glucose 6-phosphate dehydrogenase deficiency may produce bilirubin faster than it can be conjugated.

- * More bilirubin is excreted into the bile,
- The amount of urobilinogen entering the enterohepatic circulation is increased,
- Urinary urobilinogen is increased.
- Unconjugated bilirubin levels become elevated in the blood, causing jaundice.

Hepatocellular Jaundice

Damage to liver cells (for example, in patients with cirrhosis or hepatitis) can cause unconjugated bilirubin levels to increase in the blood as a result of decreased conjugation.

The bilirubin that is conjugated is not efficiently secreted into the bile, but instead diffuses (“leaks”) into the blood. Urobilinogen is increased in the urine because hepatic damage decreases the enterohepatic circulation of this compound, allowing more to enter the blood, from which it is filtered into the urine.

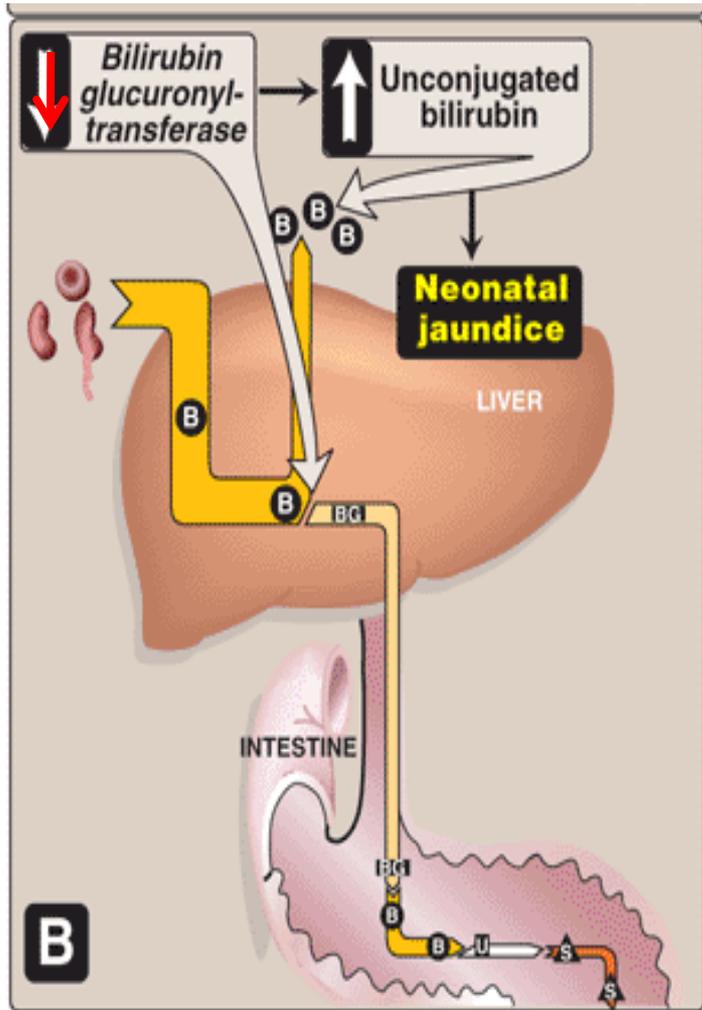
The urine thus becomes dark, whereas stools are a pale, clay color. Plasma levels of AST (SGOT) and ALT are elevated, and the patient experiences nausea and anorexia.

Obstructive Jaundice

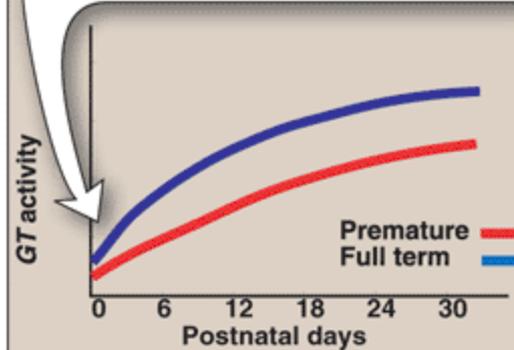
In this instance, jaundice is not caused by overproduction of bilirubin or decreased conjugation, but instead results from obstruction of the bile duct. For example, the presence of a hepatic tumor or bile stones may block the bile ducts, preventing passage of bilirubin into the intestine.

Liver regurgitate conjugated bilirubin into the blood

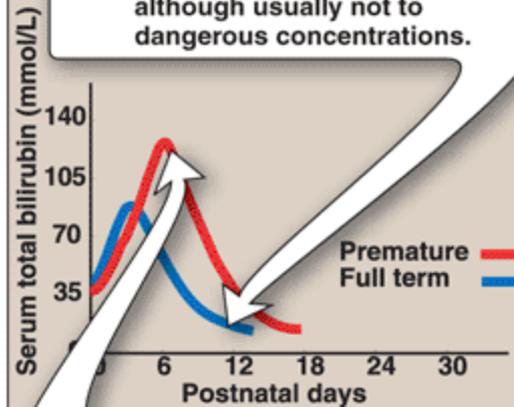
Neonatal Jaundice



1 Activity of the enzyme that conjugates bilirubin with glucuronic acid, *bilirubin glucuronyltransferase*, is low in newborns and especially low in premature babies.



2 Serum levels of bilirubin rise after birth in full-term infants, although usually not to dangerous concentrations.



3 Serum levels of bilirubin in premature infants may rise to toxic levels.

Phototherapy in neonatal jaundice



Thus, newborns with significantly elevated bilirubin levels are treated with blue fluorescent light, which converts bilirubin to more polar and, hence, water-soluble isomers. These photoisomers can be excreted into the bile without conjugation to glucuronic acid

Determination of bilirubin concentration

Bilirubin is most commonly determined by the van den Bergh reaction, in which **diazotized sulfanilic acid** reacts with bilirubin to form red azodipyrroles that are measured colorimetrically.

Only conjugated bilirubin is water soluble and reacts directly. This is called the **DIRECT bilirubin**.

To measure the unconjugated bilirubin bound to albumin, alcohol is added to release it into solution, where it can now react. This is called the **INDIRECT bilirubin**.