Metabolism of heme

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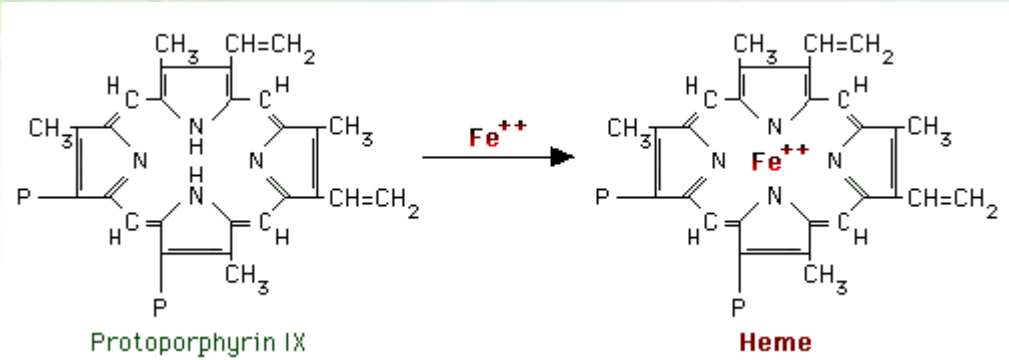
Hematopoietic-lymphatic system

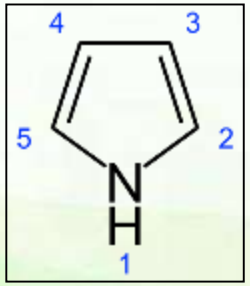
Resources

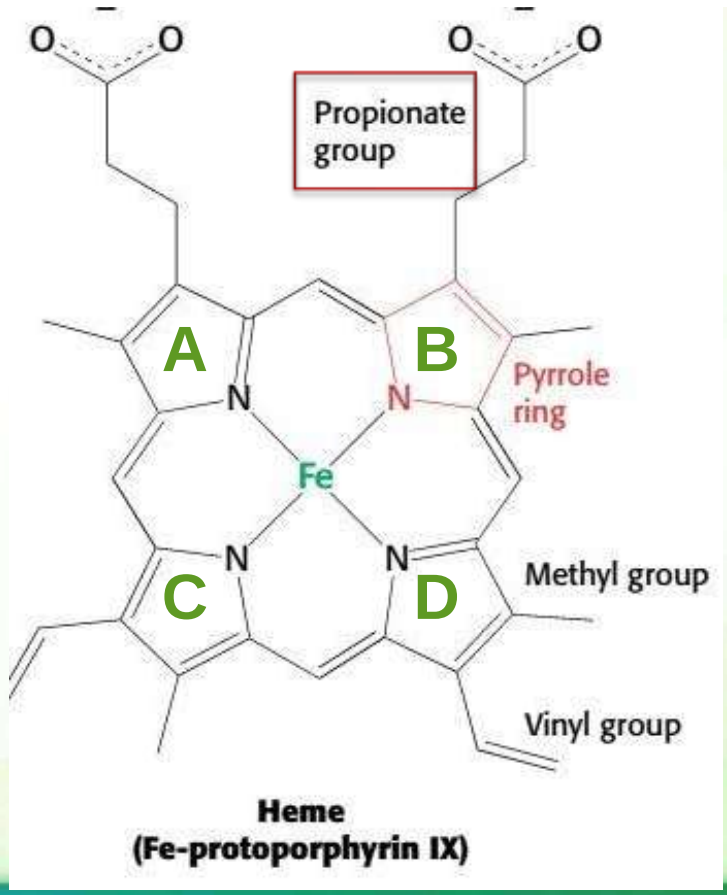
- This lecture
- Lippincott's Biochemistry, 7th edition, Ch. 21

Heme structure

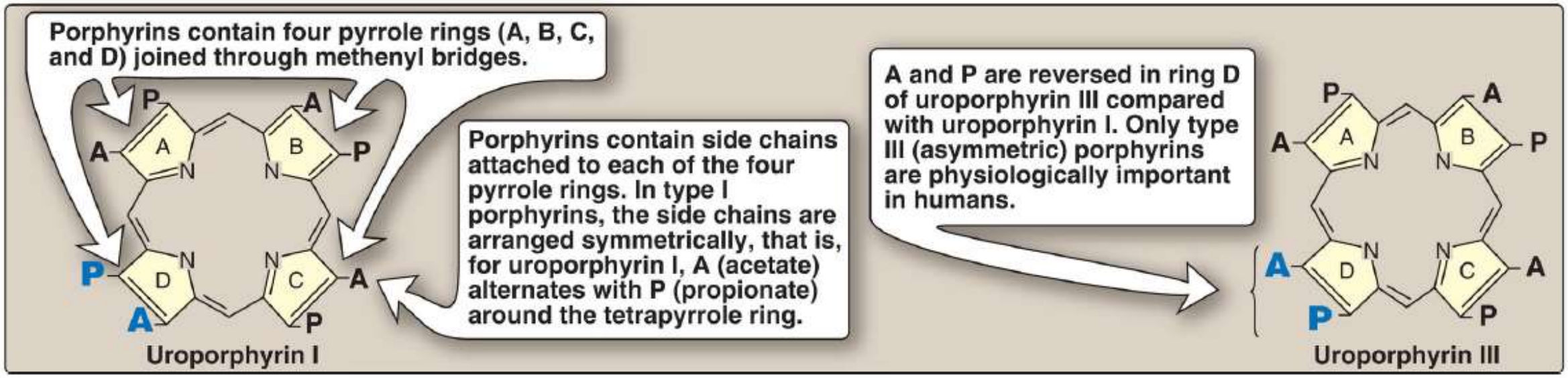
- It is a complex of protoporphyrin IX + Iron (Fe^{2+}).
- The porphyrin is planar and consists of four pyrrole rings (designated A-D).
- Each pyrrole ring can bind two substituents.
- Two rings have a propionate group each.
- Note: the molecule is hydrophobic.
- Fe has six coordinates of binding.





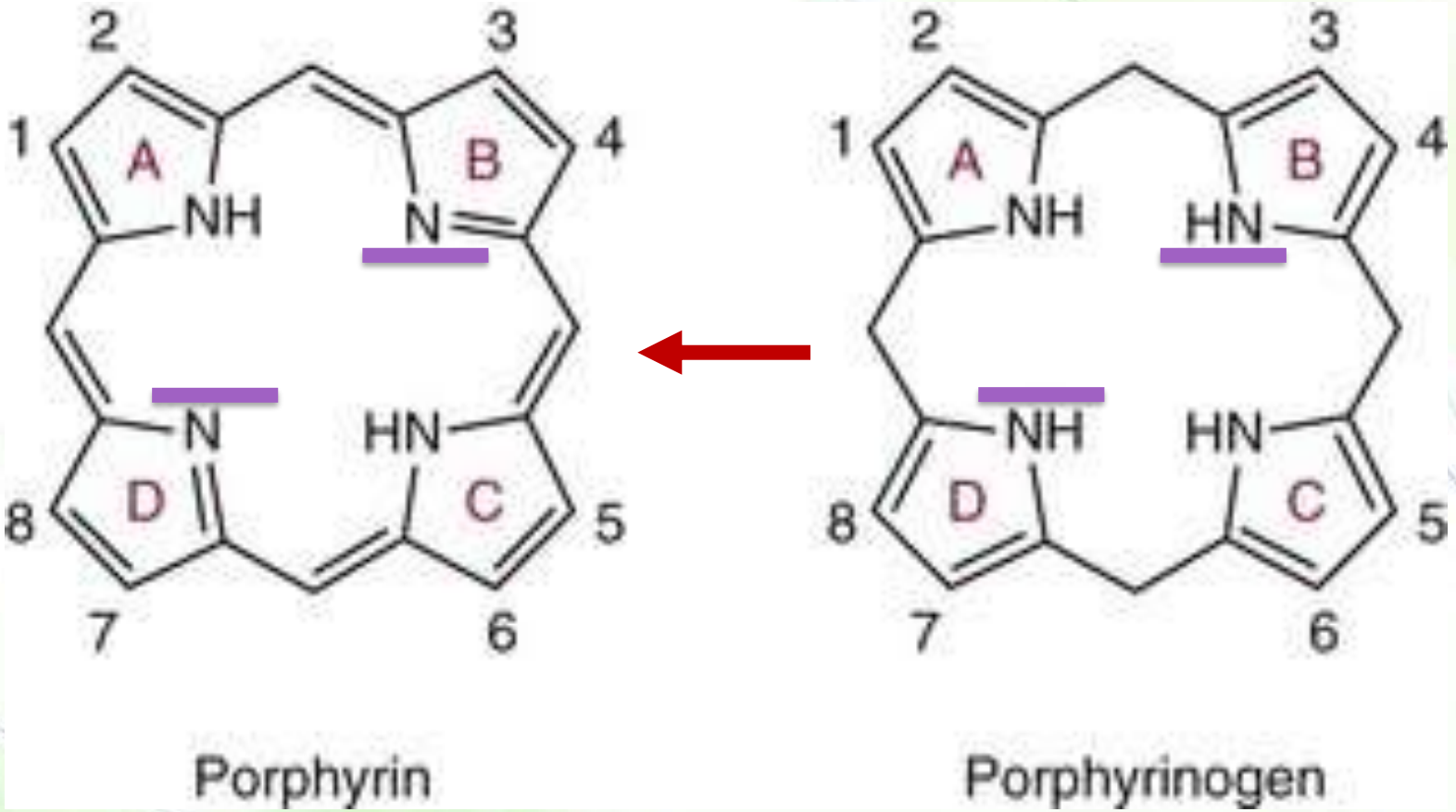


Prophyrins



Acetate **Propionate**

Porphyrinogens vs. porphyrins

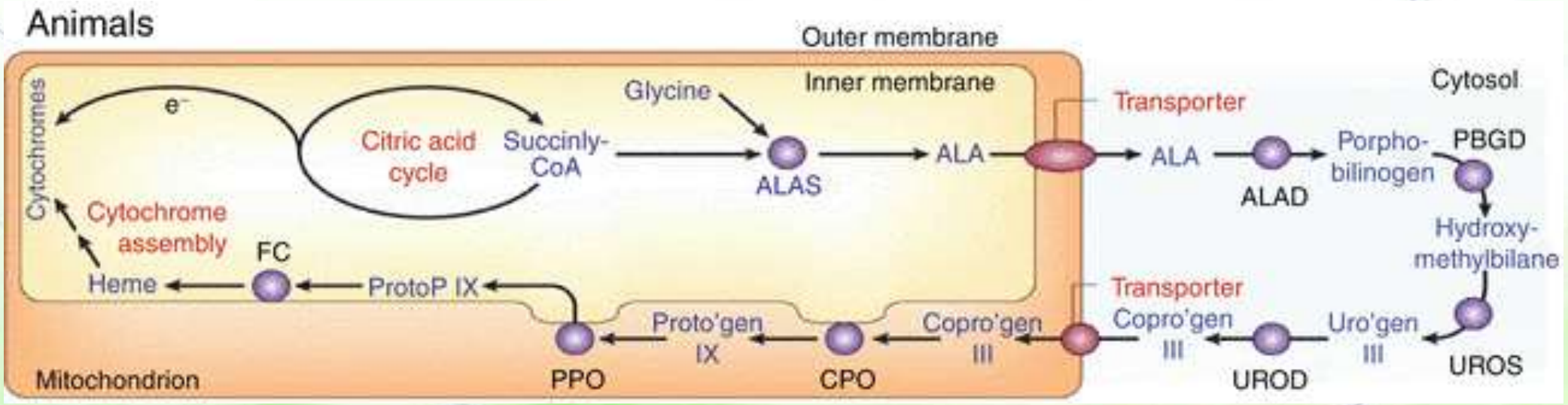


Reduced
Porphyrin precursors
Colorless
Intermediates of heme synthesis

Biosynthesis of heme

Sites of synthesis

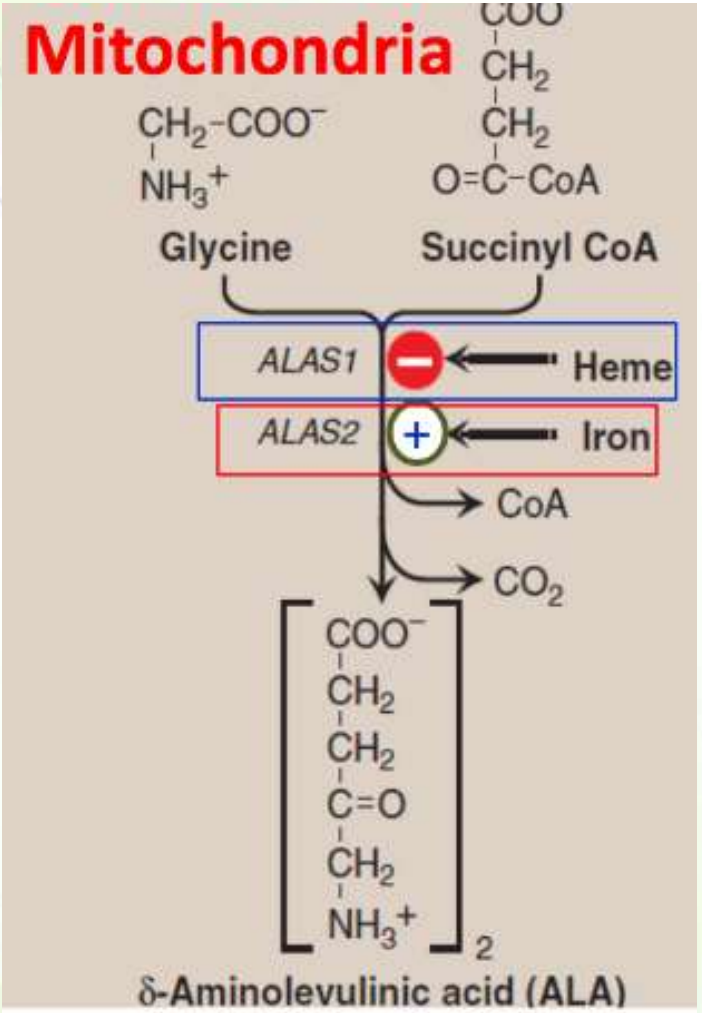
- The major sites of heme biosynthesis are:
  - Liver, which synthesizes several hemoproteins (particularly the CYP proteins)
    - The rate of heme synthesis is highly variable
  - Erythrocyte-producing cells (Hb synthesis)
    - Relatively constant production and matches the rate of globin synthesis, yet synthesis is regulated at multiple points.
- Synthesis occurs in mitochondria → cytosol → mitochondria



Synthesis of 5'-aminolevulinic acid (ALA)

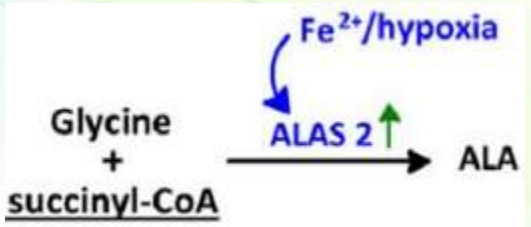
- The first reaction is catalyzed by 5’-aminolevulinic acid synthase), which conjugates glycine and succinyl CoA into ALA
  - It is the rate limiting and committed step.
  - It requires vitamin B6 (pyridoxal phosphate).
- ALA moves out of the mitochondria to the cytosol.

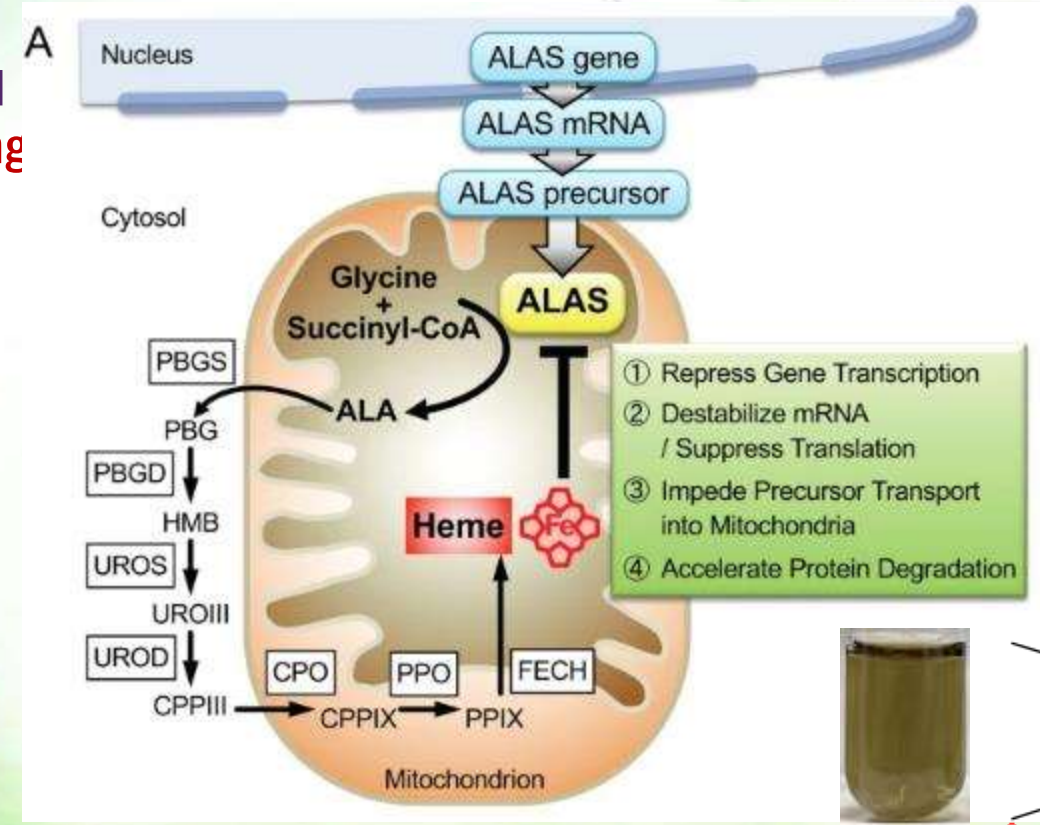
- *ALAS1 (all tissues inc. liver)*
- *ALAS2 (erythroid)*

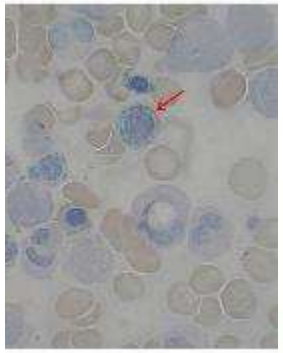


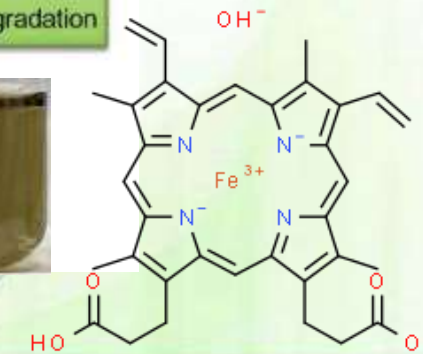
ALA synthase isoenzymes

- ALAS2 is regulated by the level of iron and hypoxia.
  - Loss-of-function mutations result in X-linked sideroblastic anemia.
    - Iron accumulates in the erythroid marrow and deposits as mitochondrial non-ferritin iron ring sideroblasts.
- ALAS1 is regulated by
  - Hemin/hematin:
    - Reduces gene transcription
    - Reduces mRNA stability
    - Inhibits mitochondrial import of ALAS1
    - Induces protein degradation
  - Drugs:







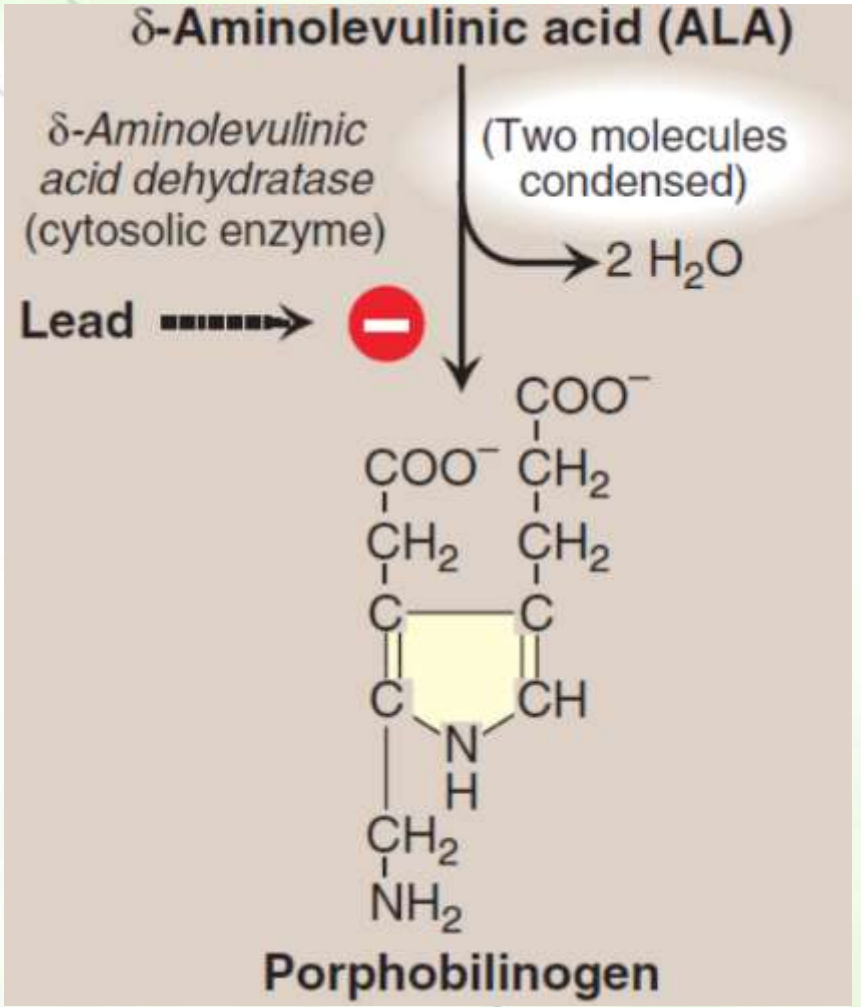


Hemin/hematin

Barbiturates → ↑CYP450 → ↓heme → ↑ALAS1 synthesis

Synthesis of porphobilinogen

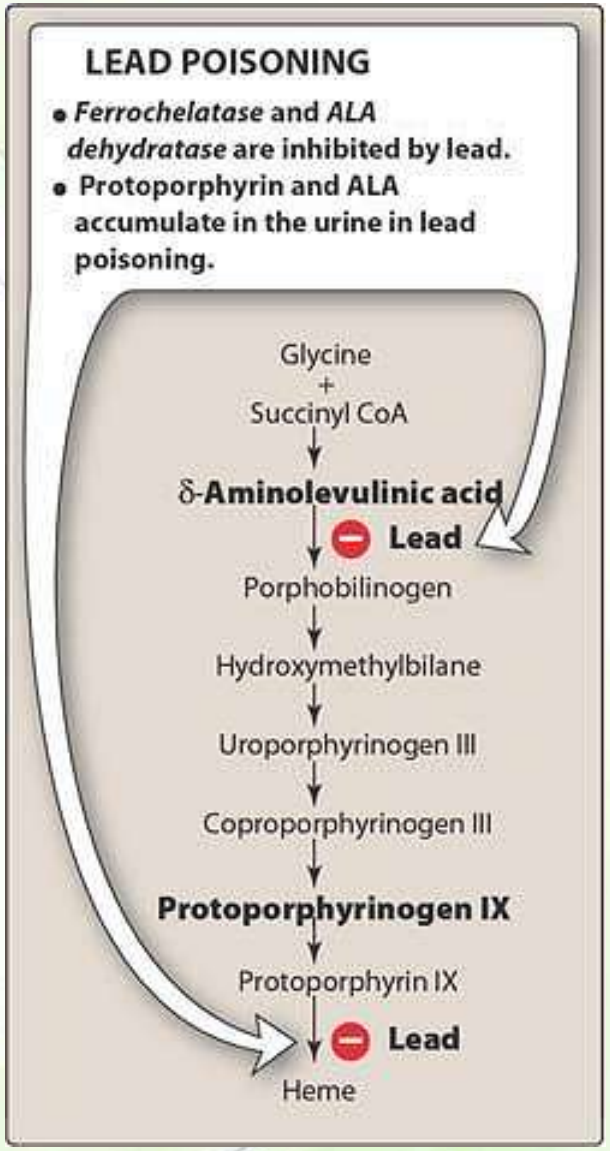
- ALA moves out of the mitochondria to the cytosol where porphobilinogen is formed by condensing 2 ALAs by zinc-containing ALA dehydratase (porphobilinogen synthase).
- The enzyme is inhibited by heavy metal ions (for example, lead) that replace the zinc.
- This inhibition causes
  - increase in ALA
  - lead poisoning-associated anemia



Subsequent reactions

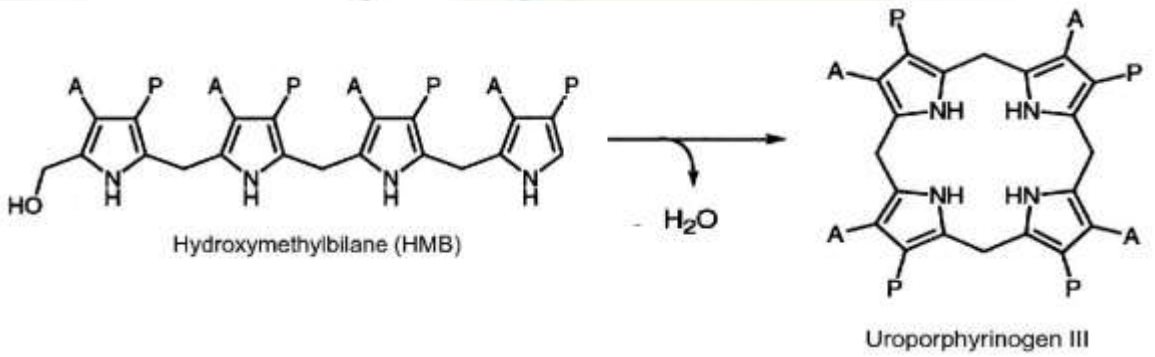
Re-record

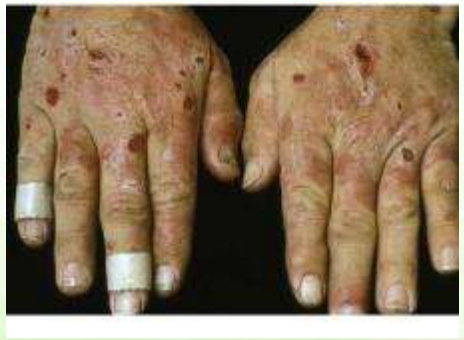
- 4x PBG → hydroxymethylbilane → *cyclic* uroporphyrinogen III → coproporphyrinogen III → *mitochondria* → protoporphyrinogen IX → *oxidized* protoporphyrin IX → (+Fe^{+2}) heme.
  - The last reaction is spontaneous but can be catalyzed by ferrochelatase.

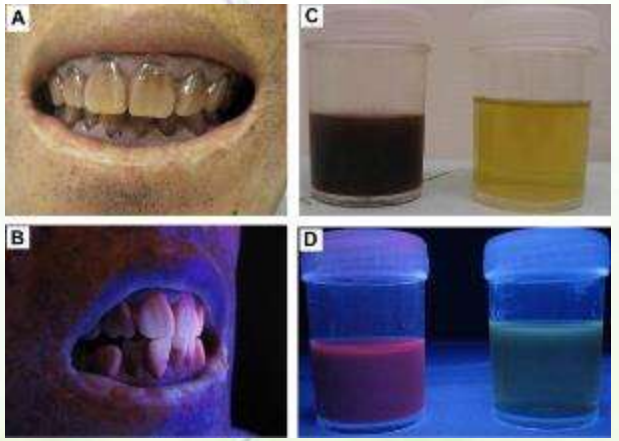


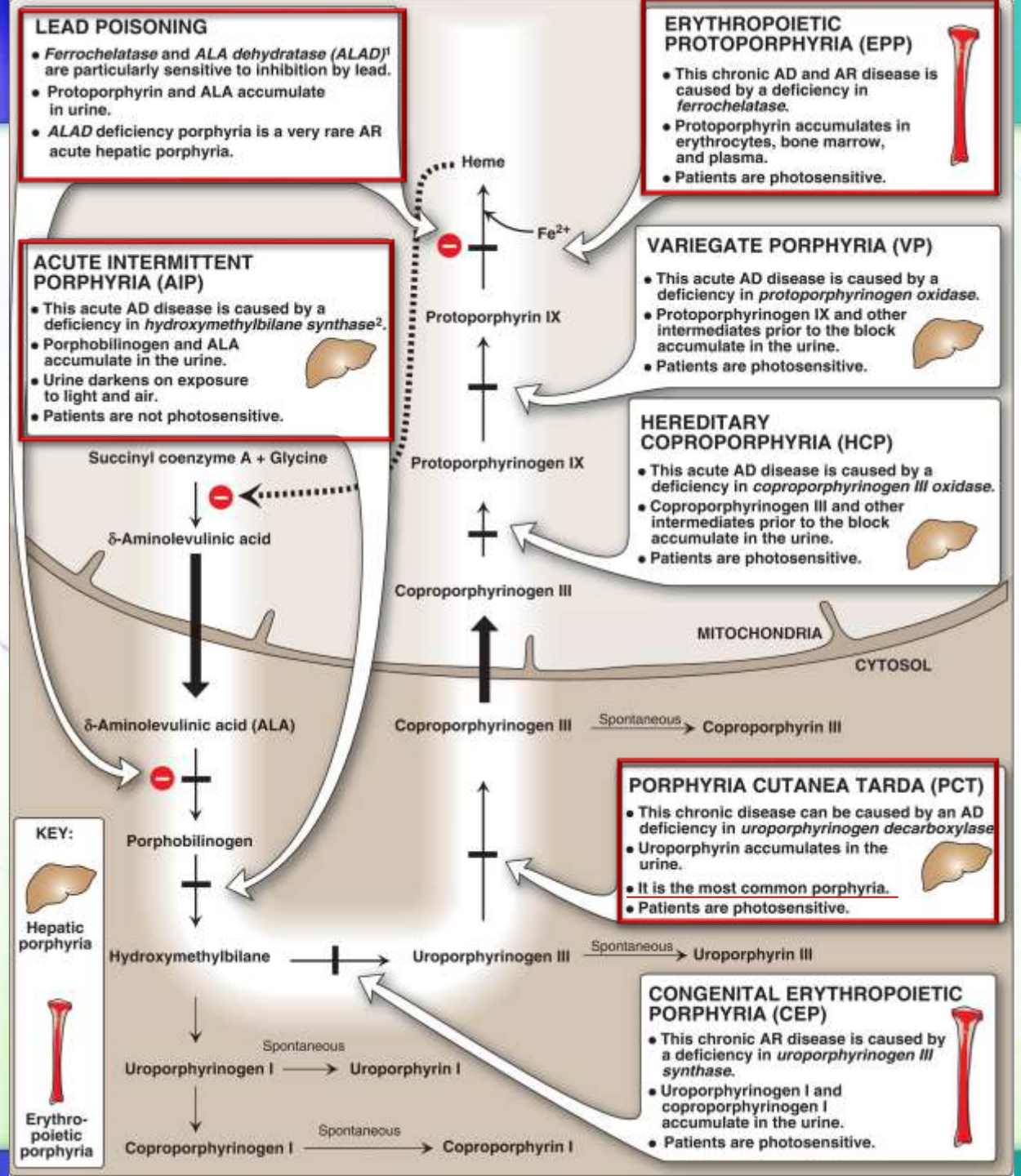
Porphyrias

- Porphyrias: inherited (mainly autosomal dominant) or acquired disorders caused by a deficiency of enzymes in the heme biosynthetic pathway resulting in elevations in the serum and urine content of intermediates in heme synthesis.
  - Porphyria = purple.
- These disorders are classified according to:
  - Affected tissue (*site of expression*):
    - Erythroid
    - Hepatic (acute or chronic)
  - Manifestations
    - Not photosensitive
      - Abdominal and neuropsychiatric
    - Photosensitive
      - Tetrapyrrole-dependent
      - Skin itching and burns
        - ↑Superoxide radicals









Hepatic porphyria

↓

↓heme

↓

↑ ALAS1 synthesis

↓

↑ intermediates

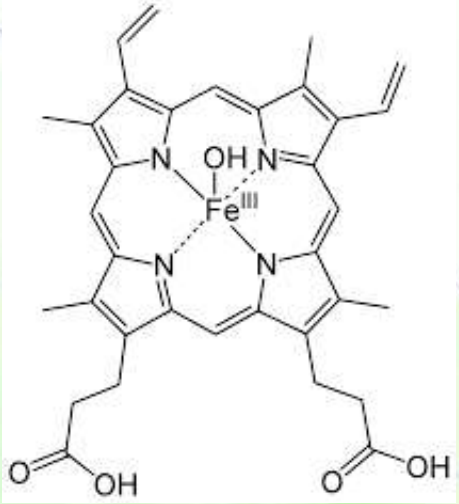
↓

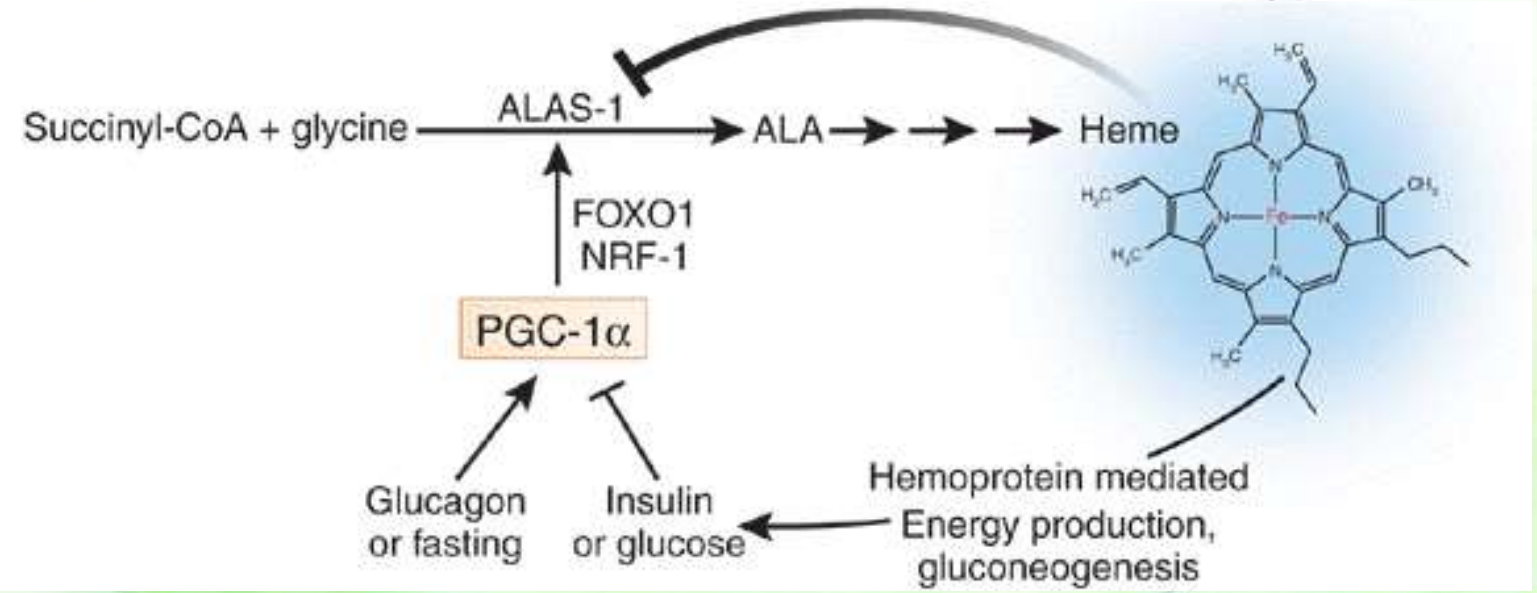
↑ toxicity

Drugs??

Treatment

- Hemin (or hematin) strongly inhibits the activity of ALAS.
- Glucose/high carbohydrate diet
  - The transcription factor, PGC-1α, is responsible for the transcription of decreases the synthesis of gluconeogenic genes and the ALAS1 gene,
    - Starvation increases the activity of PGC-1α
      - Fasting (hypoglycemia) exacerbates acute porphyria attack.
    - Feeding inhibits PGC-1α.





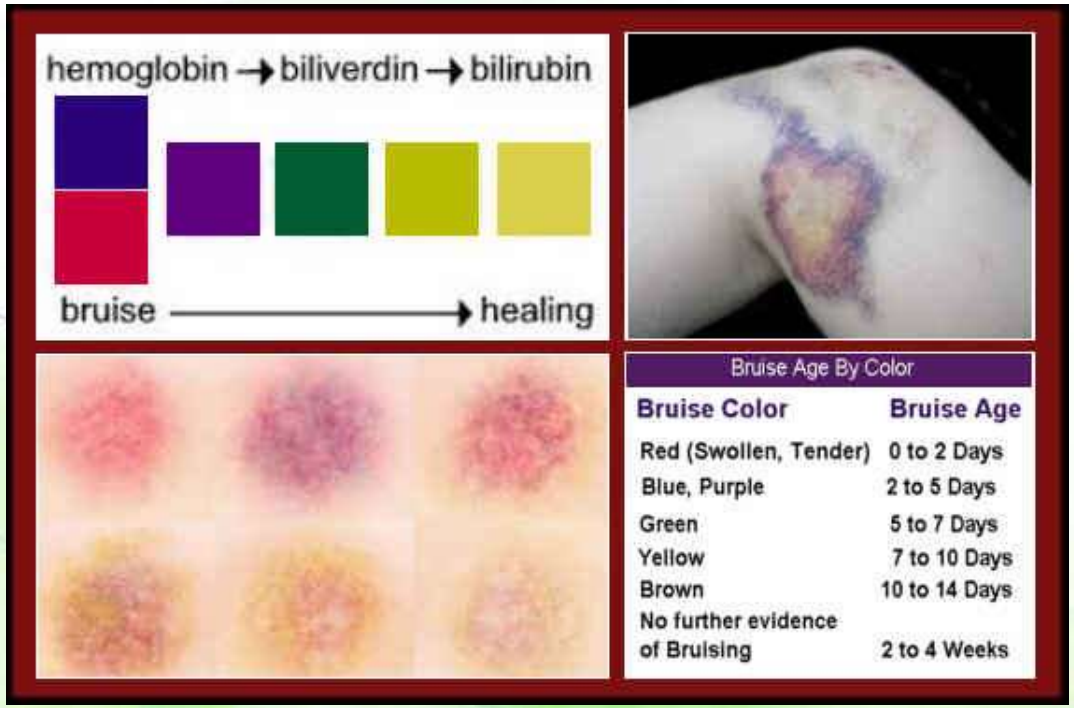
Catabolism of heme

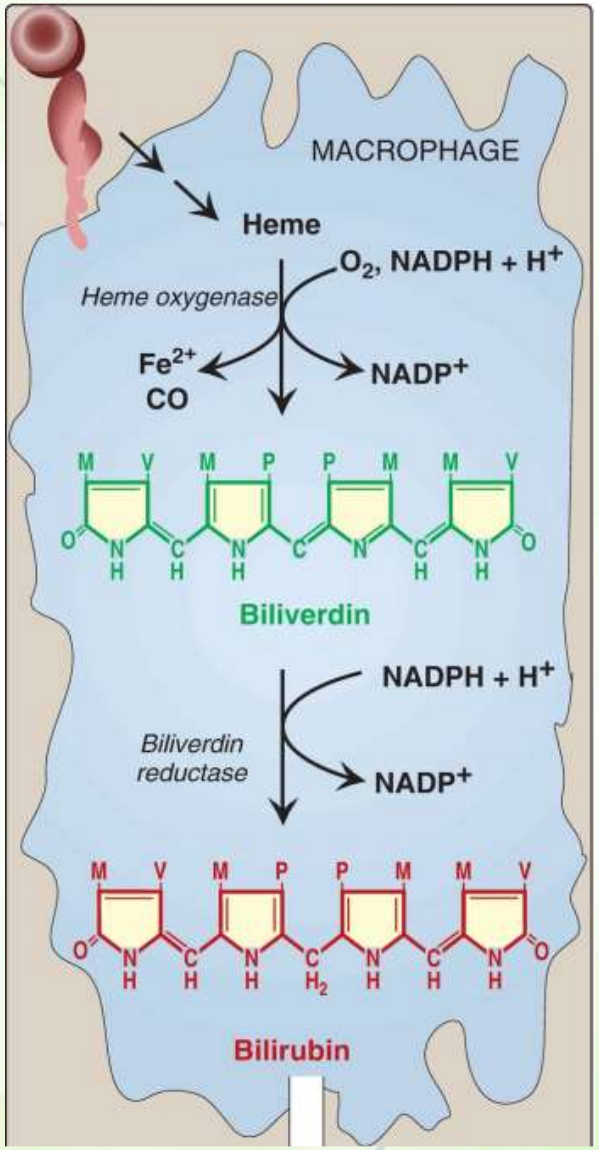
Challenges

- RBCs are the largest storage place of heme.
- Erythrocytes are mainly destroyed by macrophages in the spleen and bone marrow, releasing hemoglobin, which is degraded to heme and globin.
- The protein is metabolized into amino acids.
- 6 g/day of hemoglobin are turned over, but
  - First, the porphyrin ring is hydrophobic.
  - Second, iron must be conserved.

Heme degradation

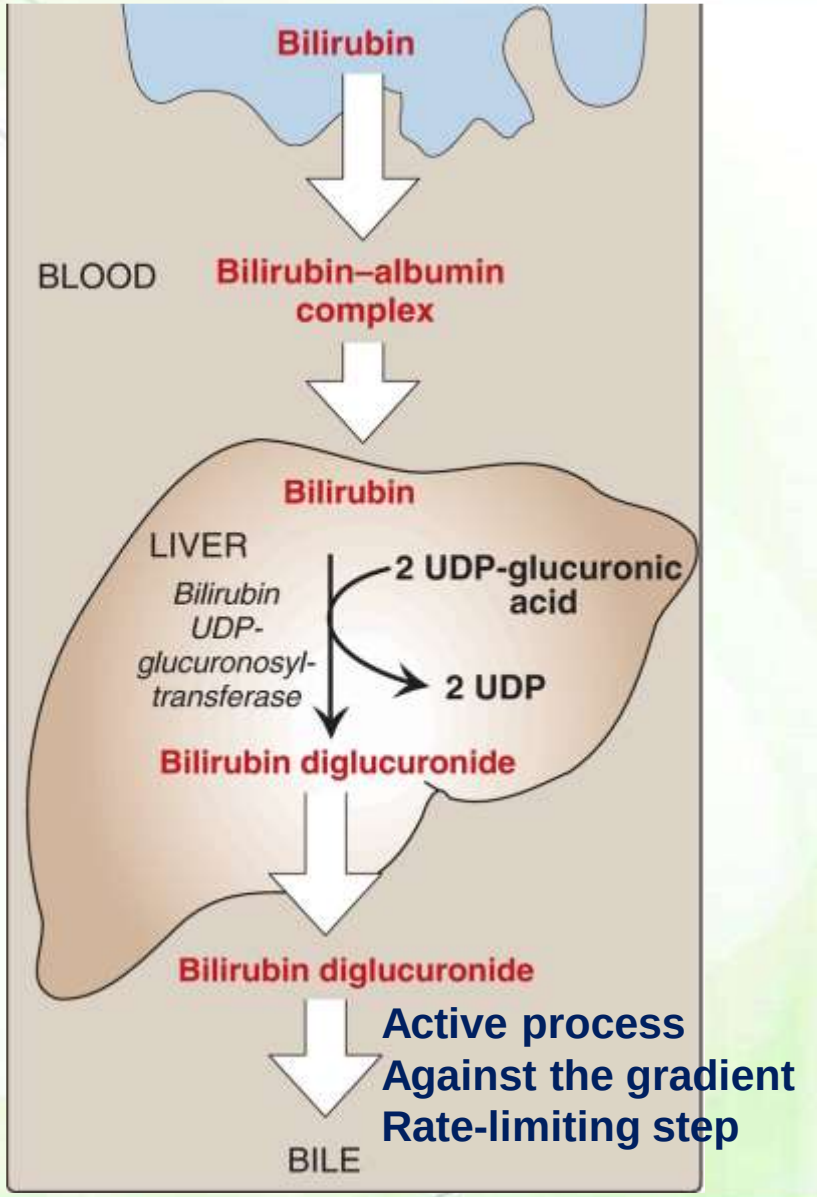
- Enzyme: heme oxygenase
  - The role of NADPH
- The production of CO
- The world of colors

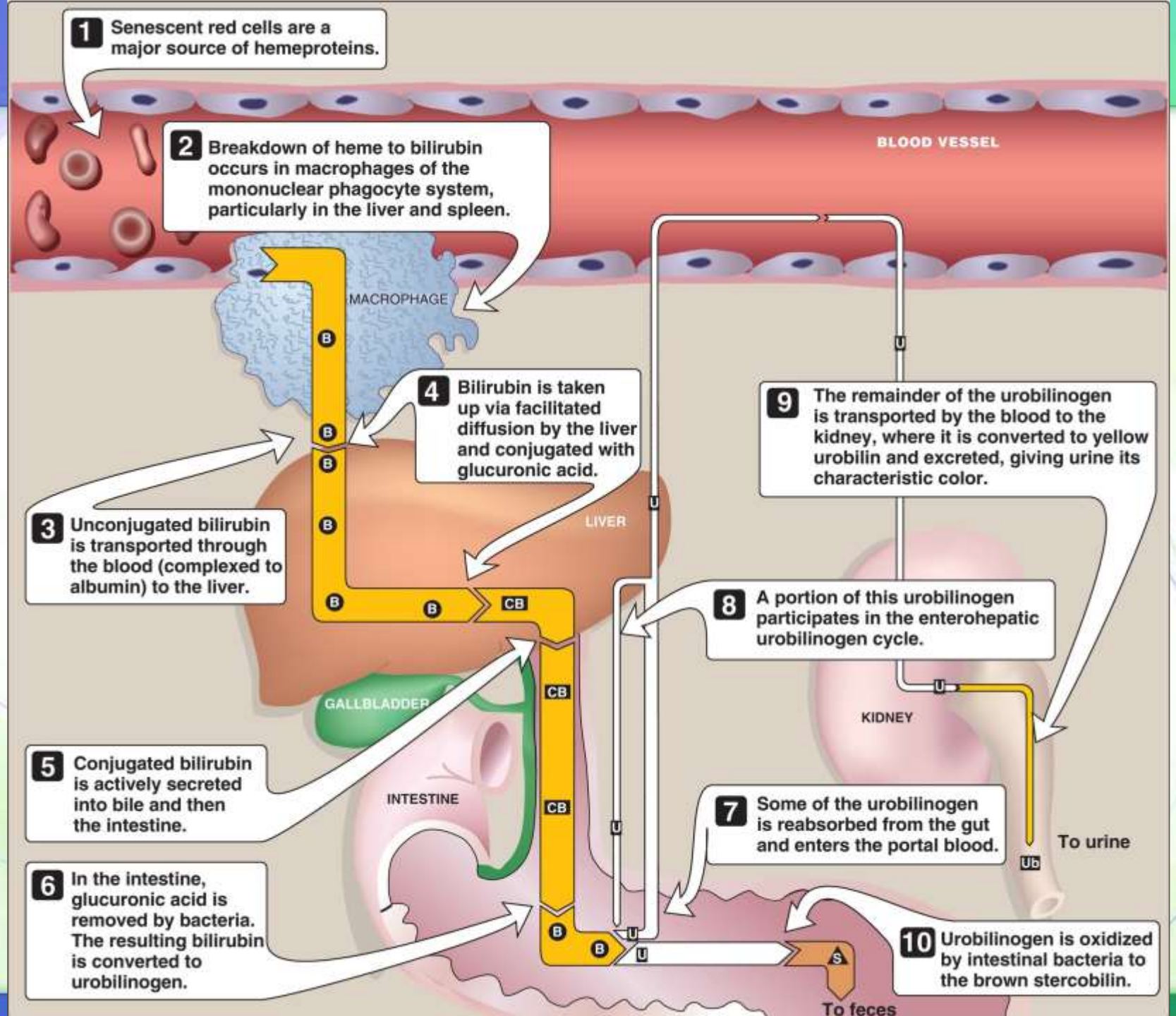




Transport of bilirubin

- The role of albumin
  - Salicylates and sulfonamides can displace bilirubin from albumin permitting bilirubin to enter the central nervous system (CNS).
    - This may cause neural damage in infants (Kernicterus).
- Formation of bilirubin diglucuronide.
  - Crigler-Najjar I and II and Gilbert syndrome
- Transport into bile
  - Dubin-Johnson syndrome

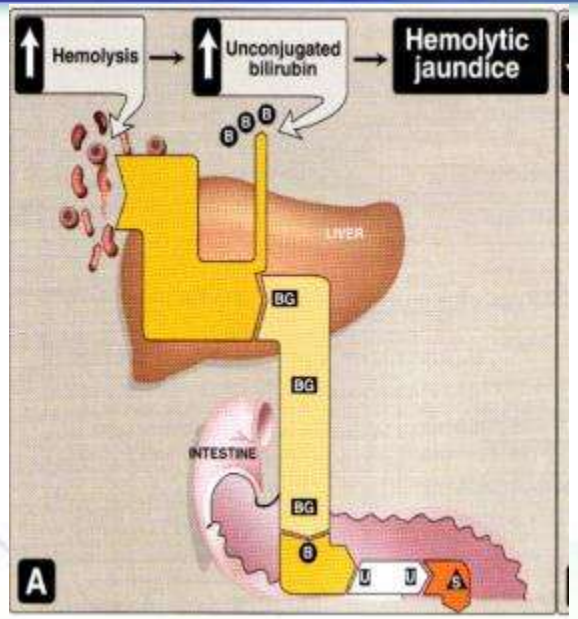


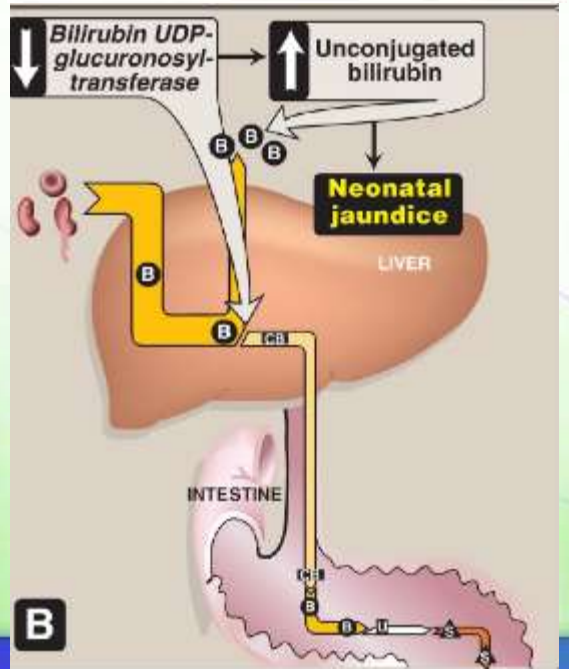

1 Senescent red cells are a major source of hemeproteins.
2 Breakdown of heme to bilirubin occurs in macrophages of the mononuclear phagocyte system, particularly in the liver and spleen.
BLOOD VESSEL
MACROPHAGE
4 Bilirubin is taken up via facilitated diffusion by the liver and conjugated with glucuronic acid.
3 Unconjugated bilirubin is transported through the blood (complexed to albumin) to the liver.
LIVER
9 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.
8 A portion of this urobilinogen participates in the enterohepatic urobilinogen cycle.
GALLBLADDER
KIDNEY
5 Conjugated bilirubin is actively secreted into bile and then the intestine.
INTESTINE
7 Some of the urobilinogen is reabsorbed from the gut and enters the portal blood.
To urine
6 In the intestine, glucuronic acid is removed by bacteria. The resulting bilirubin is converted to urobilinogen.
10 Urobilinogen is oxidized by intestinal bacteria to the brown stercobilin.
To feces
B
CB
U
Ub
S

Measurement of bilirubin

- It is done via a reaction known as Van den Bergh reaction.
- Direct measurement of conjugated bilirubin (in water)
  - Normally 4% of total bilirubin
- Total measurement of bilirubin (in ethanol or methanol)
- Indirect unconjugated bilirubin = total bilirubin – direct bilirubin

Types and lab results of jaundice

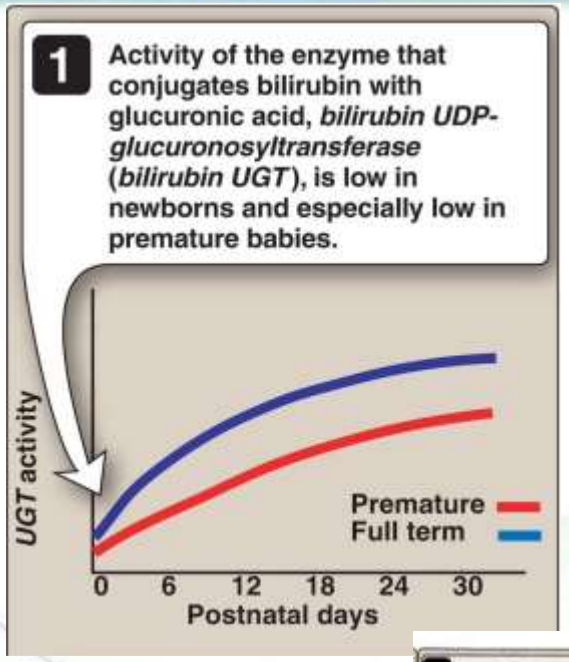


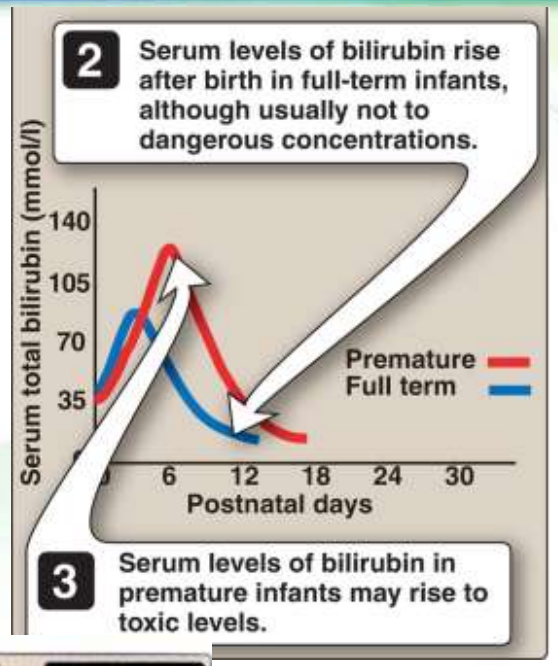


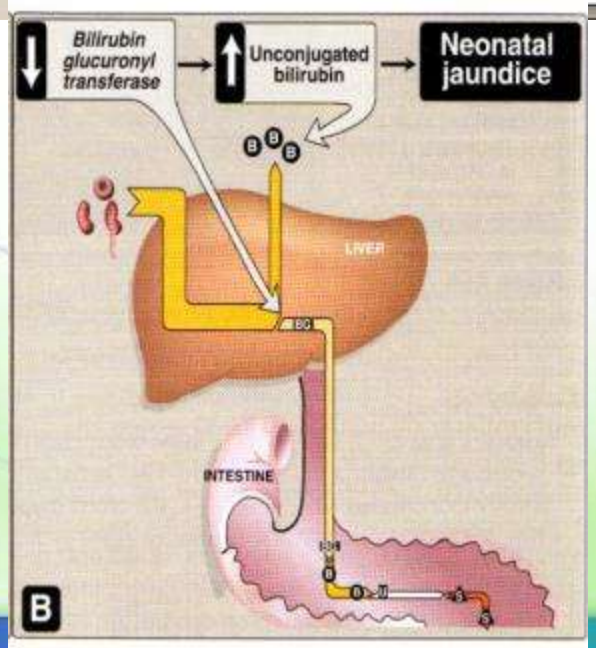
Jaundice: yellowing of skin, nail beds, and sclerae due to hyperbilirubinemia)

Sample	Indices		Unconjugated hyperbilirubinemia		Conjugated hyperbilirubinemia
		Normal	Hemolytic jaundice	Hepatic jaundice	Obstructive jaundice
Serum	Total Bil.	0.2-1.0 mg/dl	↑	↑	↔
	Direct (conj. Bil.)	0-0.2 mg/dl	↔	↑	↑
	Indirect (unconj. Bil.)	0.2-1.0 mg/dl	↑	↑	↔
	ALT/AST	Normal	Normal	↑	Normal
Urine	Color	Normal	Very dark	Dark	Dark
	Bilirubin	-	-	↑	↑
	Urobilinogen	Trace	↑	↑	↓ or -
	urobilin	Trace	↑		↓
Stool	Color	Normal	Dark	Lighter/ normal	Clayish

Jaundice in newborns







Genetic disorders

- Crigler-Najjar syndrome: severe
  - Defective conjugation by glucuronosyltransferase → Kernicterus (if type 1)
    - If type 2 (second stage conjugation) → mild
- Gilbert syndrome: defective liver uptake, mild, asymptomatic jaundice

- Defective

- Treatment:
  - Phototherapy (young age)
  - Liver transplant