Metabolism of heme

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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.
Biosynthesis of heme
The major sites of heme biosynthesis are:

- **Liver**, which synthesizes a number of 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, but synthesis is regulated at multiple points.

Synthesis occurs in mitochondria → cytosol → mitochondria
The first reaction is catalyzed by 5’-aminolevulinic acid synthase, ALAS1 (all tissues inc. liver) or ALAS2 (erythrocytes), which conjugates gly and succinyl CoA into ALA.

- It is the rate limiting and committed step.
- It requires vitamin B6 (pyridoxal phosphate).

ALA moves out of mitochondria to cytosol where porphobilinogen is formed by 2X ALA.

ALAS2 is regulated by level of iron.

ALAS1 is regulated by hemin and drugs.

- Reduced synthesis and stability of mRNA
- Inhibition of mitochondrial transport
4X PBG form uroporphobilinogen III, then coproporphyrinogen III.
Coproporphyrinogen III moves back into mitochondria.
The last reaction is spontaneous, but can be catalyzed by ferrochelatase.

In erythrocytes, synthesis is regulated at ferrochelatase and porphobilinogen deaminase (→).
1. succinyl CoA + Glycine → delta-aminolevulinic acid
2. delta-aminolevulinic acid → porphobilinogen
3. porphobilinogen → uroporphyrinogen III
4. uroporphyrinogen III → coproporphyrinogen III
5. coproporphyrinogen III → protoporphyrinogen IX
6. protoporphyrin IX → heme
Porphyrias are inherited 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 as

- Erythroid
- Hepatic (acute or chronic)

They differ in manifestations

- Photosensitive or not photosensitive
- Tetrapyrrole-dependent
- Abdominal and neuropsychiatric signs
LEAD POISONING
- Ferrochelatase and ALA dehydratase (ALAD) are particularly sensitive to inhibition by lead.
- Protoporphyrin and ALA accumulate in urine.
- ALAD deficiency porphyria is a very rare AR acute hepatic porphyria.

ACUTE INTERMITTENT PORPHYRIA (AIP)
- This acute AD disease is caused by a deficiency in hydroxymethylbilane synthase.
- Porphobilinogen and ALA accumulate in the urine.
- Urine darkens on exposure to light and air.
- Patients are not photosensitive.

ERYTHROPOIETIC PROTOPORPHYRIA (EPP)
- This chronic AD and AR disease is caused by a deficiency in ferrochelatase.
- Protoporphyrin accumulates in erythrocytes, bone marrow, and plasma.
- Patients are photosensitive.

VARIEGATE PORPHYRIA (VP)
- This acute AD disease is caused by a deficiency in protoporphyrinogen oxidase.
- Protoporphyrinogen IX and other intermediates to the block accumulate in the urine.
- Patients are photosensitive.

HEREDITARY COPROPORPHYRIA (HCP)
- This acute AD disease is caused by a deficiency in coproporphyrinogen III oxidase.
- Coproporphyrinogen III and other intermediates prior to the block accumulate in the urine.
- Patients are photosensitive.

PORPHYRIA CUTANEA TARDA (PCT)
- This chronic disease can be caused by an AD deficiency in uroporphyrinogen decarboxylase.
- Uroporphyrin accumulates in the urine.
- It is the most common porphyria.
- Patients are photosensitive.

CONGENITAL ERYTHROPOIETIC PORPHYRIA (CEP)
- This chronic AR disease is caused by a deficiency in uroporphyrinogen III synthase.
- Uroporphyrinogen I and coproporphyrinogen I accumulate in the urine.
- Patients are photosensitive.
Hemin (or hematin) strongly inhibits the activity of ALAS.

Glucose: fasting (hypoglycemia) exacerbates acute porphyria attack due to activation of the transcription factor, PGC-1α, in the liver, which induces synthesis of gluconeogenic genes and the ALAS1 gene resulting in accumulation of heme intermediates.
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.
- 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.
The roles of heme oxygenase and 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.

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.

3. Unconjugated bilirubin is transported through the blood (complexed to albumin) to the liver.

4. Bilirubin is taken up via facilitated diffusion by the liver and conjugated with glucuronic acid.

5. Conjugated bilirubin is actively secreted into bile and then the intestine.

6. In the intestine, glucuronie acid is removed by bacteria. The resulting bilirubin is converted to urobilinogen.

7. Some of the urobilinogen is reabsorbed from the gut and enters the portal blood.

8. A portion of this urobilinogen participates in the enterohepatic urobilinogen cycle.

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.

10. Urobilinogen is oxidized by intestinal bacteria to the brown stercobilin.
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

<table>
<thead>
<tr>
<th>Sample</th>
<th>Indices</th>
<th>Unconjugated hyperbilirubinemia</th>
<th>Conjugated hyperbilirubinemia</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Normal</td>
<td>Hemolytic jaundice</td>
</tr>
<tr>
<td>Serum</td>
<td>Total Bil. 0.2-1.0 mg/dl</td>
<td>↑</td>
<td>↑</td>
</tr>
<tr>
<td></td>
<td>Direct (conj. Bil.) 0.2-0.2 mg/dl</td>
<td>⇔</td>
<td>↑</td>
</tr>
<tr>
<td></td>
<td>Indirect (unconj. Bil.) 0.2-1.0 mg/dl</td>
<td>↑↑</td>
<td>↑</td>
</tr>
<tr>
<td>ALT/AST</td>
<td>Normal</td>
<td>Normal</td>
<td>↑</td>
</tr>
<tr>
<td>Urine</td>
<td>Color Normal</td>
<td>Darker</td>
<td>Dark</td>
</tr>
<tr>
<td></td>
<td>Bilirubin -</td>
<td>Present</td>
<td>Present</td>
</tr>
<tr>
<td></td>
<td>Urobilinogen Trace</td>
<td>↑</td>
<td>↓ or -</td>
</tr>
<tr>
<td></td>
<td>urobilin Trace</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>Stool</td>
<td>Color Normal</td>
<td>Dark</td>
<td>Lighter/normal</td>
</tr>
</tbody>
</table>
Jaundice in newborns

1. Activity of the enzyme that conjugates bilirubin with glucuronic acid, bilirubin UDP-glucuronosyltransferase (bilirubin UGT), 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.