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Lecturer in Toxicology

METHEMOGLOBIN-FORMING AGENTS

1. Definition of MetHb

Methemoglobin (MetHb) is a non-functional hemoglobin in which iron changes from the ferrous state (Fe^{2+}) to the ferric state (Fe^{3+}), which is unable to bind oxygen (O_2).

2. Physicochemical properties of MetHb

1. **Spectral properties:** it absorbs in the visible range, which allows its measurement.
2. It is attacked by strong acids and bases (degraded into amino acids).
3. MetHb combines with:
    - CN^-: cyanmethemoglobin
    - H_2S: sulfmethemoglobin
    - F^-: fluoromethemoglobin
    - NO_2: nitrosomethemoglobin

3. Physiological formation of MetHb

It is continuously formed inside red blood cells. This oxidation is due to normal metabolism (physiological), and the MetHb level normally remains below 2% because it is rapidly reduced by methemoglobin reductase I.

Normal levels (physiological):
Adult: 0.5–0.8%
Newborn: 1.5%
Premature: 2%

4. Methemoglobin-forming agents

Classification

<u>According to chemical nature</u>

1. Mineral compounds (direct action):

Nitrites (NO_2^-), chlorates, $KMnO_4$

-Nitrites

Origin:

- Endogenous: synthesized from the amino acid arginine
- Exogenous: drinking water, animal products, vegetables, fertilizers
  Note: nitrites result from the reduction of nitrates by bacteria

Toxic dose:

Maximum allowed concentration in drinking water = 45 mg/L

Toxicity:

Methemoglobinemia, dangerous for young children

-Chlorates ($NaClO_3$ / $KClO_3$)

Origin: herbicide, oral solutions, dyes, explosives, match heads

Toxic dose:

Ingestion of less than 20 matches is not dangerous for a child

Lethal dose: $NaClO_3$ = 15 g, $KClO_3$ = 7 g

Toxicity:

Severe methemoglobinemia with hemolysis and anuric tubulopathy

Digestive causticity: nausea, vomiting, epigastric pain

-$KMnO_4$

Use: dermatological antiseptic

Toxicity:

Severe methemoglobinemia with hemolysis

Digestive causticity: nausea, vomiting, oropharyngeal pain

2. Organic compounds (indirect action):

Aromatic nitro and amino derivatives, methylene blue (MB: both MetHb-forming agent and antidote), antileprosy drugs (dapsone), primaquine, benzocaine, poppers.

<u>According to mechanism of action</u>

- **Direct methemoglobin-forming agents:** high redox potential, act directly on hemoglobin producing superoxide and H_2O_2
  Example: ferricyanide/ferrocyanide, hydrazines, nitrites, chlorates

- **Indirect agents:** metabolized into active compounds

  Example: nitrobenzene

5. Mechanism of action

Fe^{2+} forms 6 coordination bonds:

- 4 with tetrapyrrole nitrogen
- 1 with globin histidine
- 1 with O_2

Fe^{3+} forms 6 coordination bonds:

- 4 with tetrapyrrole nitrogen
- 1 with globin histidine
- 1 with H_2O → oxygen binding becomes impossible

Consequences of oxidation

- Hemoglobin oxidation → MetHb formation
- Lipid peroxidation by ROS → membrane lysis
- Hemoglobin denaturation → Heinz bodies formation, membrane binding, cell lysis and hypoxia

Consequences of MetHb in blood

- Brown chocolate coloration of blood
- Free MetHb loses heme → binds albumin → methemalbumin → brown plasma

6. Reduction mechanisms of MetHb

- **NADH-dependent pathway (main):** reduces 2/3 of MetHb
- **NADPH-dependent pathway:** requires electron carrier (MB)
- **Glutathione pathway:** secondary (10–15%)
- **Ascorbic acid pathway:** weak, effective if MetHb <15%

7. Factors influencing MetHb formation

- Alcohol: increases oxidation, produces ROS
- Fetal hemoglobin: more sensitive
- Digestive disorders: increased sensitivity
- G6PD deficiency: decreased reduction pathways
- Hepatic and renal insufficiency

8. Clinical features of poisoning

As MetHb increases → cyanosis

- **>10%:** cyanosis (bluish nails, lips, mucosa)
- **>20%:** fatigue, headache, dizziness, tachycardia, polypnea, chocolate-colored blood
- **>60%:** CNS damage, coma, respiratory depression, cardiac arrest
- **>70%:** death

9. Treatment

Symptomatic: oxygen therapy, anticonvulsants

Decontamination:

- Skin washing
- Gastric lavage
- Nasal irrigation

Elimination:

- Exchange transfusion if severe

Antidotes

1. Methylene blue (MB)

- Mechanism: electron donor → reduces MetHb
- Indication: MetHb > 40%
- Contraindications: G6PD deficiency
- Administration: IV slow
- Side effects: anxiety, dyspnea, hypertension, cyanosis

2. Ascorbic acid (Vitamin C)

- Mechanism: reduction of MetHb
- Less effective but non-toxic
- Used in mild cases

10. Toxicological analysis

Sample: blood (without NaF)

Methods:

- Spectrophotometry (Kaplan method, Evelyn-Malloy)
- CO-oximeter