Haemolytic anaemias

Dr. J Potgieter
Department of Haematology
NHLS Tshwane Academic Division
Red Cell Destruction

- Extravascular in macrophages of RES (BM, liver, spleen)
- Haem to iron and protoporphyrin
- Globin to amino acids
- Hb binds to haptoglobin and the complex is removed by the liver. Haptoglobin is produced by the liver – low levels in haemolysis and liver disease
- Pathological red cell destruction may occur intravascular
Clinical Features

- Anaemia (unless compensated)
- Jaundice (mild)
- Splenomegaly (many types)
- Ankle ulcers (e.g. sickle cell anaemia)
- Pigment gallstones
- Bone expansion in children due to marrow expansion (e.g. frontal bossing)
- Aplastic crises (Parvo virus or folate deficiency)
Lab Features

- Normal or reduced Hb
- Biochemistry: raised unconjugated bilirubin, low haptoglobin, increased f-stercobilinogen & u-urobilinogen, raised LDH
- Increased reticulocyte count
- Bone marrow: hyperplastic erythropoiesis
- Blood film: polychromasia, normoblasts. red cell shape changes (e.g. schistocytes, spherocytes)
Lab Features

- **Radioactive chromium labeling of red cells**
  - Measures lifespan
  - Assess sites of destruction
- **Intravascular haemolysis**
  - Increased plasma & urine Hb
  - Methaemalbumin
  - Urine-haemosiderin
Causes of intravascular haemolysis

- Mismatched blood transfusions
- G6PD-deficiency with oxidant stress
- Red cell fragmentation syndromes
- Some auto-immune haemolytic anaemias
- Some drug and infection induced haemolytic anaemias
- Paroxysmal Nocturnal Haemoglobinuria
- March haemoglobinuria
- Unstable haemoglobins
# Classification of haemolytic anaemia

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<td>Metabolism</td>
<td>Warm antibody type: Idiopathic/secondary</td>
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<tr>
<td>G6PD deficiency</td>
<td>Cold antibody type: Idiopathic/secondary</td>
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<td>Haemoglobin</td>
<td>Alloimmune</td>
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<td>HbS, HbC</td>
<td>Haemolytic transfusion reaction</td>
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Inherited Haemolytic Anaemias
Membrane Abnormalities

1. Hereditary spherocytosis
   - Whites, AD (variable severity); 25% new mutation
   - Deficient/abnormal RBC membrane proteins (vertical)
   - Lipid bilayer unsupported causing loss of lipid/membrane passing through RES (esp. spleen) – low surface area:volume
   - Spherocytes less deformable and osmotically fragile
   - $K^+$ loss and dehydration
Clinical features

- Mild anaemia, splenomegaly, mild jaundice
- Severe transfusion-dependent anaemia
- Aplastic crises
- Increased haemolysis during infection
- Gall stones (50%)
- Lower leg ulcers
- Iron overload
Laboratory features

- Hb varies
- Micropspherocytes and polychromasia (reticulocytosis), occasional normoblasts
- MCHC increased (50%)
- Increased osmotic fragility (median corpuscular fragility –MCF) and auto-haemolysis
- Negative direct antiglobulin test
- Red cell membrane protein analysis (SDS-PAGE, sodium dodecyl sulphate polyacrylamide gel electrophoresis)
Typical Graphs for RBC Osmotic Fragility

Hemolysis

100%
80%
60%
40%
20%
0%

0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9

% NaCl Solution

Sickle Cell Anemia
Thalassemia Major

Normal Range

Hereditary Spherocytosis
- Treatment
  - Folic acid
  - Blood transfusion
  - Splenectomy
  - Cholecystectomy
Enzyme abnormalities

1. Glucose 6 phosphate dehydrogenase (G6PD) deficiency
   - Deficiency renders cells susceptible to oxidant stress (e.g. drugs, infection, fava beans etc.)
   - Inheritance is sex-linked
   - Blacks, Mediterranean, middle eastern, oriental
   - Protection against malaria
Hexose monophosphate pathway

Clinical picture

- Haemolysis 1-3 days after drug exposure or febrile illness or hours to days after ingestion of Fava beans
- Severe cases: abdominal/ back pain, dark urine
- Heinz bodies appear and Hb decreases rapidly
- Neonatal jaundice (impaired conjugation) esp. Gilbert disease
- Hereditary nonsheroctic haemolytic anaemia (chr jaundice, splenomegalcy, gall stones)
Laboratory features

- Normal blood film between crises
- Bite / blister cells and polychromasia with acute intravascular haemolysis. Heinz bodies on supravital staining
- Screening test (fluorescent spot test) for generation of NADPH in steady state
- Enzyme assays and DNA analysis

Management
- Stop offending drug, treat infection, transfuse red cells, consider splenectomy
1. Thalassaemia

- Commonest single gene disorders
- Mediterranean region hence thalassaemia (greek Θαλασσα, meaning ‘the sea’)
- Autosomal recessive inheritance
- α or β depending on reduced synthesis of α or β globin chains
The genetics of α-thalassaemia

- Normal (αα/αα); silent carrier (α-/αα)
- α-Thal trait (α-/α-) or (--/αα). Low MCV, MCH
- HbH disease (--/α-). Hypochrome, anaemia, splenomegaly, HbH
- Hydrops fetalis (--/--)

Phenotype

- **B thalassaemia (β^0/β^+)**
  - Minor: symptomless carrier state
  - Intermedia: anaemia and splenomegaly but does not require regular transfusions
  - Major: severe, transfusion dependent
Clinical features

- Anaemia (3-6 months), milder cases later ≤ 4 years
- Failure to thrive, intercurrent infection, mild jaundice
- HSM, expansion of bones
- Iron overload
Laboratory features

- Blood film: Hypochromic microcytic anaemia, target cells and erythroblasts
- Raised RCC >5.5 \( \times 10^{12}/l \)
- HPLC
- Hb electrophoresis:
  - B thalassaemia maj: HbF 10-90%, HbA absent if \( \beta^0 \) thal.
  - B thalassaemia min: HbA2 > 3.5%
- Hypercellular bone marrow (erythroid hyperplasia)
- Globin chain synthesis studies
- DNA analysis shows specific mutations
Haemoglobin electrophoresis/
HPLC

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<th>ANALYTE ID</th>
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TOTAL AREA: 1121158

F: 56.0%  A2: 2.3%
Management

- Maintain Hb > 9 – 10 g/dl
- Iron chelation therapy
- Immunise: hep B
- Splenectomy
- Bone marrow transplant
Autoimmune haemolytic anaemia (AIHA)
Warm-type
- Red cells coated with IgG alone or complement
- 37\(^\circ\)C
- Taken up by the RE macrophages
- Part of coated membrane is lost → progressively more spherical
Warm-type AIHA

- **Idiopathic**
  - 30%
  - Any age (infancy, early childhood, 3rd decade, > 5th decade)
  - Evans syndrome
  - AIHA of infancy and childhood

- **Secondary**
  - SLE, other autoimmune diseases
  - CLL, lymphomas
  - Drugs (e.g. fludarabine, methyldopa)
  - Malignancies
  - Viral infections
Clinical and Lab features

- Pallor or jaundice
- Splenomegaly (2-3 cm under costal margin)
- Blood film: microspherocytes, polychromasia, $\pm$ normoblasts
- Direct antiglobulin test positive
- IgG or IgG and complement
Treatment

- Corticosteroids (1-2mg/kg/d)
- Cytotoxic immunosuppressive drugs
  - Azathioprine (1.5-2 mg/kg/d)
  - Cyclophosphamide (1.5-2mg/kg/d)
- Cyclosporin (5mg/kg/d in 2 divided doses)
- IVIG (0.4mg/kg/d X 4-5 days)
- Splenectomy
- Blood transfusion
Cold-type AIHA

- Idiopathic cold haemagglutinin disease
  - Chronic course in older people
  - Acrocyanosis in cold weather
  - Spontaneous agglutination of red cells
  - DAT shows C3d

- Secondary
  - Infections: mycoplasma pneumonia, infectious mononucleosis
  - Lymphoma

- Paroxysmal cold haemoglobinuria
Cold-type AIHA

- Ab whether monoclonal (ICHID) or polyclonal (infection) attaches to membrane in cooler peripheral circulation
- Usually IgM and binds best at 4°C
- Fixes complement and extra- and intravascular haemolysis can occur
- Spherocytes less marked, agglutinated red cells
Treatment

- General: management is difficult, avoid exposure to cold, folic acid supplements
- Alkylating agents: Chlorambucil intermittent or continuous
- Corticosteroids and splenectomy are rarely of use
- Blood transfusion and plasma exchange
Drug induced immune haemolytic anaemia

- Ab against drug-red cell membrane complex. Drug acts as hapten (penicillin)
- Ab against drug-plasma protein complex → subsequent deposition of immune complexes on red cells (quinidine, rifampicin)
- Stimulation of auto Ab against red cells (methyldopa, fludarabine)
Red cell fragmentation syndromes

- Physical damage to red cells on abnormal surfaces or fibrin strands deposited in small vessels
- Termed microangiopathic haemolytic anaemia (TTP/HUS, pre-eclampsia, meningococcal septicaemia, adeno Ca)
- Haemolysis intra and extravascular
- Fragmented red cells
- Arterial grafts, cardiac valves
- Disseminated intravascular coagulation (DIC)
- Thrombotic thrombocytopenic purpura (TTP)
- Haemolytic uraemic syndrome (HUS)
- Meningococcal sepsis
- Pre-eclampsia and HELLP syndrome
Infections

- Direct damage to red cells (malaria)
- Toxin production (clostridium)
- Oxidant stress (G6PD- deficiency)
- MAHA (meningococcal septicaemia)
- Antibody formation (infectious mononucleosis)
- Extravascular destruction (malaria)
Chemical and physical agents

- Certain drugs → oxidative intravascular haemolysis (dapsooon, salazopyrin)
- Wilson’s disease → acute haemolysis due to high levels of copper in the blood
- Chemical poisoning with lead, arsine
- Severe burns damages red cells
Paroxysmal nocturnal haemoglobinuria

- Clonal disorder, acquired mutation of X-chromosome gene coding for phosphatidyl inositol glycan A (PIG-A)
- Result = Glucosyl Phosphatidyl Inositol (GPI)-linked proteins are absent from the red cell e.g:

  Decay-activating factor (DAF, CD55)
  Membrane inhibitor of reactive lysis (MIRL, CD59)
PNH – general features

- Red cells are sensitive to lysis with complement causing chronic intravascular haemolysis
- Haemosiderinuria leads to iron deficiency
- Recurrent venous thrombosis and infections are prevalent
- The marrow may be hypoplastic with neutropenia and thrombocytopenia
Diagnosis

- Positive acid lysis (Ham’s) test
- Absence of CD55 and CD59 antigens

Management

- Supportive (folic acid, iron if deficient, blood transfusions, short course steroids, oral anticoagulation)
- BM transplant and immunosuppressive R
References
