Hemolytic Anemias

Objectives:
1. Explain the role of immune reactions, red blood cell membrane defects, red blood cell enzyme defects, or hemoglobin abnormalities in the development of hemolytic anemia.
2. List the characteristic findings of hemolytic anemia.
3. Describe the presentation and management of Glucose 6 Phosphate Dehydrogenase deficiency hemolytic anemia.
4. Describe the presentation of thalassemia major.
5. Describe the complications of sickle cell disease.

Hemolytic Anemia
Describes a group of anemia of that are all characterized by abnormal destruction of red cells. The hallmark of these disorders is reduced life span of the red cells rather than underproduction by the bone marrow. The red cell normally survives about 120 days, but in hemolysis the cell survival times are considerably shortened.

Sites of hemolysis
I. Intravascular hemolysis:
   When red cells are rapidly destroyed within the circulation, hemoglobin is liberated, e.g.;
   1. Glucose-6-phosphate dehydrogenase (G6PD) deficiency,
   2. Hemolytic Uremic Syndrome/thrombotic thrombocytopenic purpura,
   3. Disseminated intravascular coagulation,
   4. Following transfusion of ABO incompatible blood,
   5. Hemolytic anemia due to prosthetic cardiac valves and
   6. Paroxysmal nocturnal hemoglobinuria (PNH)

II. Extravascular hemolysis:
The red cells are removed from the circulation by macrophages in the reticuloendothelial system, particularly the liver and spleen. e.g.
   1. Autoimmune hemolytic anemia and
   2. Hereditary spherocytosis.

III. Intramedullary hemolysis:
Hemolysis may also be intramedullary when fragile RBC precursors are destroyed in the bone marrow prior to release into the circulation. e.g. thalassemia major.
**Laboratory findings**

I. Features of increased RBC breakdown (intra or extra vascular hemolysis)
   1. Increased unconjugated serum bilirubin
   2. Increased urine urobilinogen
   3. Increased faecal stercobilinogen
   4. Absent serum haptoglobin, because it becomes saturated with Hb.

II. Features of increased RBC production
   1. Reticulocytosis
   2. Bone marrow erythroid hyperplasia

III. Damaged RBC
   1. Morphology; microspherocytes, elliptocytes, fragments
   2. Osmotic fragility, autohemolysis
   3. Shortened RBC survival

**Classification of Hemolytic Anemia**

I. Inherited disorders
   1. RBC membrane defect; Hereditary spherocytosis and Hereditary elliptocytosis
   2. Enzyme defect; Glucose $-6$-phosphate dehydrogenase and Pyruvate kinase deficiency
   3. Hemoglobin defects; Thalassemia syndromes and Sickling disorders

II. Acquired disorders
   1. Immune mediated;
      i. Auto immune (autoimmune hemolytic anemia),
      ii. Alloimmune (hemolytic disease of newborn, hemolytic transfusion reaction)
   2. Non-immune and trauma; valve prosthesis, microangiopathy, infection, drugs or chemicals, hypersplenism, secondary (liver and renal disease)

**Hemolytic anemias = reduced red-cell life span**

*Cardinal features; pallor - jaundice - splenomegaly*

*Diagnosis of hemolytic syndrome:*
   1. Anemia
   2. Reticulocytosis
   3. Indirect hyperbilirubinemia
**Red cell enzyme defect**

**G6PD Deficiency**
Most common RBC enzyme defect; it is seen in 3% of population
This hemolytic anemia results from oxidative damage to RBCs as a consequence of the loss of the protective effect of the enzyme G6PD. Cells from G6PD-deficient subjects can’t convert the oxidized substrate to a reduced state.

**Genetics**
G6PD deficiency is sex linked, affecting males & carried by females who show half normal RBC G6PD values.
There are many variants: A+, A-, B+, B-.
B -; the most common variant in Mediterraneans (which is associated with potentially life-threatening hemolytic disease because of severe deficiency).

**Clinical presentations:**
The most common clinical feature of glucose-6-phosphatase dehydrogenase (G6PD) deficiency is a lack of symptoms (ie; Persons with this condition do not display any signs of the disease until their red blood cells are exposed to certain chemicals in food or medicine or to stress).

I. **Neonatal jaundice:** Jaundice usually appears by age 1-4 days
II. **Acute hemolytic anemia beyond the neonatal period:** Clinical expression results from exposure to stress factors such as oxidative drugs or chemicals, infection, or ingestion of fava beans.
   - Rapidly developing intravascular hemolysis: Jaundice, dark urine and pallor.
   - The anemia may be self-limiting as new young RBCs have near normal enzyme levels.
   - Outside of these episodes; no evidence of chronic hemolysis exists.

Typically; most patients who have G6PD deficiency are asymptomatic until exposed to an inciting agent, at which time they may develop acute hemolysis. The onset of hemolysis usually is within 24 to 48 hours of exposure. Initial manifestations may include acute abdominal pain, vomiting or diarrhea, low-grade fever, and hemoglobinuria (cola-colored urine), followed by the appearance of jaundice and symptoms of anemia such as lethargy and irritability.

Physical examination usually reveals anemia, jaundice, splenomegaly, and hepatomegaly are unusual.
In severe cases, cardiovascular decompensation may occur.

III. **Chronic hemolysis (congenital nonspherocytic hemolytic anemia)**
• **Hallmark of hemolysis;** *(Anemia, Reticulocytosis, Indirect hyperbilirubinemia)*

**Diagnosis of G6PD deficiency:**

I. During an attack, the following lab. findings are present:

1. Anemia
2. Peripheral Blood film (smear) shows;
   - Normochromic normocytic anemia of varying degrees
   - Small cells (poikilocytes), some of which are spherocytic or fragmented.
   - Characteristic findings include “bite” cells, which are RBCs with areas that are bitten off; Spleen removes portion of RBC that had Heinz body, preventing intravascular hemolysis.
   - The WBC count usually is elevated as a result of hyperactivity of the marrow, but the platelet count may be normal, elevated, or reduced.
3. Reticulocytosis
4. Heinz bodies (inclusion bodies) detected by using special (supravital) stains; these bodies are denatured hemoglobin found attached to the cell membrane from the interior of the cell (May damage cell membrane, leading to intravascular hemolysis).
5. There is evidence of intravascular hemolysis;
   - Serum haptoglobin is reduced,
   - Increased unconjugated hyperbilirubinemia,
   - In severe cases, hemoglobinemia and hemoglobinuria.

II. G-6-PD enzyme deficiency can be detected using several screening tests. A high reticulocyte count may mask this (during the acute attack) as young red cells have higher G6PD activity. Therefore, the best time to perform this test is several weeks following hemolysis, specifically when the reticulocyte count has normalized.

III. The blood count is normal between attacks

**Treatment**

- During the attack:
  1. Remove offending drug & treat any underlying infection
  2. Brisk hydration to ensure adequate urine output that will prevent clogging of renal tubules.
  3. Transfusion may be necessary in severe hemolysis (Obtain blood sample before transfusion)
- Teaching to avoid oxidant drugs, chemicals, & fava beans
- Genetic consultation, evaluation of family members

**Course of the disease:**

- Most acute hemolytic episodes are mild and self-limiting, even in the face of continuing exposure, because there is an increased level of enzymes in the remaining younger population of cells, anemia usually resolves in 3 to 6 weeks.
- Possible Complication; rare renal failure or death may occur following a severe hemolytic event.

**The differential diagnosis:**

1. Autoimmune hemolytic anemia,
2. Hemolytic -uremic syndrome, and
3. Malaria-induced hemolysis.

**Prognosis:** Spontaneous recovery from a hemolytic episode is the usual outcome.

**Auto Immune hemolytic anemia (AIHA)**

AIHA is characterized by the production of antibodies against an individual’s own erythrocyte membrane antigens, which leads to hemolysis. It is an acquired form of hemolysis with a defect arising outside the red cell. The bone marrow produces structurally normal red cells and premature destruction is caused by the production of an aberrant autoantibody targeted against one or more antigens on the cell membrane. The disease can be divided into ‘warm’ and ‘cold’ types depending on whether the antibody reacts better with red cells at 37°C or 4°C.

The peak incidence occurs in the preschool age group. The hemolysis in AIHA is largely extravascular, usually involves IgG, and occurs primarily in the liver and spleen.

**Clinical features**

Patients who have AIHA usually present with pallor, jaundice, lethargy, abdominal pain, or low-grade fever. If hemolysis is severe, the urine may be dark. Among the signs are those associated with hyperdynamic circulation, including an enlarged spleen and liver.

**Laboratory findings**

I. Anemia; Hemoglobin level—very low

II. Peripheral blood smear; prominent spherocytes, polychromasia, macrocytes, autoagglutination (antibodies best detected at 37°C)

III. Reticulocytosis—common

IV. Evidence of hemolysis:
   1. Hyperbilirubinemia
   2. Haptoglobin level—markedly decreased
   3. Hemoglobinuria
   4. increased urinary urobilinogen

V. Direct coomb’s test—positive

**Treatment**

*Most patients who have AIHA exhibit mild anemia of limited duration and, therefore, do not need therapy.* Because most chronic cases are mild, minimal or no intervention is necessary. If treatment is necessary, several modalities are known to be effective.

I. Compensated hemolysis – observation (clinical evaluation) and folic acid at an oral dose 1mg/day

II. Decompensated hemolysis (definitive therapy depends on the cause):
   - Steroid therapy; Prednisolone 2mg/kg up to 6mg/kg & should then be tapered down
   - Immunoglobulins
   - Splenectomy can be helpful where extravascular haemolysis is predominant (ie most of the red blood cells are being removed by the spleen).
   - Blood transfusion: This treatment approach is needed only in life-threatening situations or as a temporary solution while waiting for other modalities to begin working. The “least incompatible” blood may be the best choice because of cross-matching difficulties.
**Thalassemia syndromes**
The thalassemias are inherited disorders of Hb synthesis that result from an alteration in the rate of globin chain production. A decrease in the rate of production of a certain globin chain or chains (alpha, beta, or delta) impedes Hb synthesis and creates an imbalance with the other, normally produced globin chains. Their clinical severity widely varies, ranging from asymptomatic forms to severe or even fatal entities.

**B-Thalassemia:**
There are two beta genes.

I. Beta thalassemia trait/thalassemia minor; Deficiency of one beta genes leads to essentially no significant hemolysis and no unusual signs or symptoms are encountered.

II. Thalassemia major; Deficiency of both genes leads to significant hemolytic anemia.

III. Thalassemia intermedia is a condition in which the degree of hemolysis is milder even though the patient may have a deficiency of both beta genes. Therefore, thalassemia intermedia is essentially a descriptive term that refers to minimal or no need for transfusions.

**ß-thalassaemia major**

**Clinical features:**
- Anemia first becomes apparent between 3-6 months when production of HbF declines, the infant clinically normal at birth (as fetal Hb does not contain ß chains) then during first year of life; Progressive anemia will occur.
- Failure to thrive
- Hepatosplenomegaly & jaundice.
- The severity of this anemia results from a combination of ineffective erythropoiesis and shortened survival of the red blood cell in circulation.

**Complication:**
- Complications of **hemolytic anemia** include expansion of bones leads to thinning of cortex & tendency to fractures, bossing of skull, and specific facies with hair-on-end appearance on x-ray, gallstones, and chronic leg ulcers.
- **Iron overload** caused by repeated transfusion, increased iron absorption due to ineffective erythropoiesis; The complications of iron overload affect all organs of the body, including the heart (arrhythmias, heart failure), liver (cirrhosis), thyroid (hypothyroidism), pancreas (diabetes), and hypothalamic-pituitary axis (delayed growth and sexual maturity) unless chelation therapy is given.
- **Infections** secondary to splenectomy & blood transfusion transmitted viruses
Laboratory diagnosis
The CBC count and peripheral blood film examination results are usually sufficient to suspect the diagnosis.
Hemoglobin (Hb) evaluation confirms the diagnosis in β thalassemia

I. Complete blood picture:
   1. In the severe forms of thalassemia, the Hb level ranges from 2-8 g/dL.
   3. Blood film shows nucleated red cells, target cells, polychromasia, punctate basophilia and anisopoikilocytosis, basophilic stippling
   4. Mean corpuscular volume (MCV) and mean corpuscular Hb (MCH) are significantly low.

II. Hemoglobin electrophoresis: elevated HbF (>50%) with variable A2.

III. Evidence of hemolysis: Unconjugated hyperbilirubinaemia.

IV. Iron studies show increased iron stores as follows:
   1. High serum iron level
   2. High serum ferritin level

V. DNA analysis.
   This test is used to investigate deletions and mutations in the beta globin producing genes. Family studies can be done to evaluate carrier status and the types of mutations present in other family members.

VI. Both parents will have β-thalassemia trait

Management
I. Blood transfusion: these children are entirely transfusion dependent, to maintain the Hb > 10g/dL, by transfusion every 4-6 weeks with fresh, filtered blood

II. Iron chelation: start after 10-15 units of blood,
   1. Desferioxamine 20-40 mg/kg by s.c infusion over 8-12 hours, 5-7 days weekly,
   2. Deferasirox (Exjade) a new oral chelator with few side effects is a promising drug

III. Splenectomy; indicated if the spleen is enlarged substantially or there is evidence of hypersplenism with or without a massive size leading to pancytopenia Can reduce the transfusion frequency.

IV. Stem cell transplantation (SCT): In best risk patients the probability of survival exceeds 90%.

V. Gene therapy: Stimulation of fetal hemoglobin production and somatic gene therapy
Sickle Cell Disease
Are groups of Hemoglobinopathies, which primarily affect the Afro-Caribbean population.
The common feature of these diseases is inheritance of an abnormal hemoglobin β chain gene—the gene is designated β S.
HbS differs from normal Hb (HbA) in that glutamic acid has been replaced by valine at the sixth amino acid from the N-terminus of the B globin chain.

Pathogenesis:
• In HbS, valine is substituted for glutamic acid in the sixth amino acid of the β chain.
• Deoxy-HbS is much less soluble than deoxy HbA; it forms a gelatinous network of fibrous polymers that cause RBCs to sickle at sites of low pO₂.
• Hemolysis - because sickle RBCs are too fragile to withstand the mechanical trauma of circulation
• Occlusion in microvascular circulation caused by distorted, inflexible RBCs adhering to vascular endothelium

Clinical features:
The clinical features of sickle cell anemia arise from the propensity of red cells containing haemoglobin S to undergo ‘sickling’. In the deoxygenated state; the HbS molecules aggregate into long polymers which then align to form liquid crystals (tactoids). The red cell loses its normal deformability and becomes characteristically sickle shaped.
The variation ranges from a complete lack of symptoms even into adulthood to life-threatening complications beginning in early infancy
• Most affected patients, however, have long periods of “wellness” interspersed with recurrent symptomatic episodes
• VASO-OCLUSION EPISODES: : The pathogenesis of pain is disruption of blood flow in the microvasculature by sickle cells, resulting in tissue ischemia and often accompanied by pain, can affect nearly all organs or tissues (except probably the myocardium). The most common sites are the bones, lungs, spleen, liver, penis, and brain. Precipitating events can include infection, stress, dehydration, or changes in temperature.
• PAINFUL BONE EPISODE; This is the most frequent vaso-occlusive episode and results from infarctions of the bone marrow and cortical bone. It may occur early in the first year of life as “dactylitis” that involves the small bones of the hands and feet (hand-foot syndrome), but more commonly it occurs
later. No bone in the body is spared, but the spine, ribs, and long bones are affected most frequently. Clinical features include pain of acute or gradual onset with or without signs of local inflammation.

- **TRANSIENT RED BLOOD CELL APLASTIC EPISODE;** Acute infection with parvovirus B19 is usually associated with red cell aplasia, fever, and pain.

- **ACUTE SEQUESTRATION EPISODE;** This is an acute trapping of blood in the spleen and less frequently in the liver caused by impaired egress of blood out of these organs due to clogging by sickled cells.

- **INFECTIONS;** Infections are common among patients who have sickle cell disease because functional asplenia which can appear as early as 6 mo of age in some children and by 5 yr of age in most children, it results from multiple infarcts leads to impaired production of antibodies and opsonins and the absence of the splenic function of phagocytosis of bacterial organisms. The risk of infection is highest with encapsulated organisms such as pneumococci, *Haemophilus influenzae*, meningococci, and *Salmonella* sp.

- **Others;** Gall stones, chronic osteomyelitis, chronic leg ulcers, Lung disease (acute chest syndrome), Renal disease, Recurrent priapism, etc

**Diagnosis of SCA**

2. Screening tests for sickling: the blood sample is deoxygenated to induce sickling.
3. Hemoglobin electrophoresis: in sickle cell anemia (HbSS)

**Treatment**

I. Treatment of vaso-occlusive crisis include:
   1. Hydration
   2. Pain control,
   3. Empirical antibiotics
   4. Blood transfusion (simple or exchange transfusion)

II. Chronic transfusion therapy.

III. Medical intervention; Stimulation of increased hemoglobin F production (hydroxyurea),

IV. cellular and genetic levels include
   1. Stem cell transplant, and
   2. Gene therapy

V. Children with sickle cell anemia should receive prophylactic oral penicillin at least until 5 yr of age

VI. Routine childhood immunizations and annual administration of influenza vaccine are highly recommended.