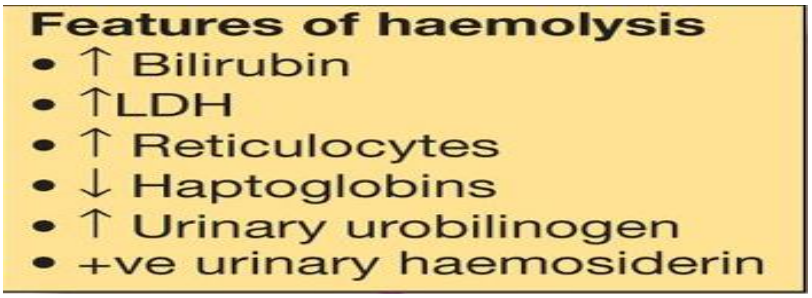
Hemolytic anemias

Hemolytic anemia represents approximately 5% of all anemias. Hemolysis is the premature destruction of erythrocytes.The normal red cell lifespan of 120 days may be shortened by a variety of abnormalities. The bone marrow may increase its output of red cells six- to eight-fold by increasing the proportion of red cells produced, expanding the volume of active marrow and releasing reticulocytes prematurely. If the rate of destruction exceeds this increased production rate, then anemia will develop

Hemoglobin breakdown, causing rise in unconjugated bilirubin in the blood and mild jaundice. Increased reabsorption of urobilinogen from the gut results in an increase in urinary urobilinogen. Red cell destruction releases LDH into the serum.

Reticulocytosis, and nucleated red cell precursors also appear in the blood, Activation of the bone marrow can result in a neutrophilia and immature granulocytes appearing in the blood to cause a leuco-erythroblastic blood film.



Blood film in hemolytic anemia :

- Spherocytes :Small round cells with no central pallor Hereditary spherocytosis; immune hemolytic anemia.
- Schistocytes (Fragmented RBCs): occur in Mechanical trauma: malfunctioning prosthetic heart valve; thrombotic microangiopathy (TTP/HUS); severe burns; severe shock or acidosis; severe intravascular hemolytic anemia
- Sickle cells: Curved RBCs with pointed ends as in Sickle cell anemia and other sickle cell disease
- Target cells : Central thick area surrounded by pale ring as in Liver disease; hemoglobinopathies (thalassemia, sickle cell anemia).
- Polychromasia : RBCs with bluish color on routine stains Young RBCs (reticulocytes); often seen in hemolytic anemias or recovery from blood loss.
- Heinz body : Blue dots in RBC; seen in Unstable hemoglobin; Unstable hemoglobins; hemolytic anemia due to enzyme deficiencies (G6PD deficiency)

Intravascular hemolysis:

free hemoglobin is released into the plasma. Free hemoglobin is toxic to cells and binding proteins have evolved to minimize this risk(haptoglobin)

Haptoglobin is an α_2-globulin produced by the liver which binds free hemoglobin, resulting in a fall in levels of haptoglobin. Once haptoglobins are saturated, free hemoglobin is oxidized to form methemoglobin which binds to albumin, in turn forming methaemalbumin. Methaemoglobin is degraded and any free hem is bound to a second binding protein termed hemopexin.

free hemoglobin may appear in the urine. When fulminant, this gives rise to black urine, as in severe falciparum malaria infection. In smaller amounts, renal tubular cells absorb the hemoglobin, degrade it and store the iron

as hemosiderin. When the tubular cells are subsequently sloughed into the urine, they give rise to hemosiderinuria, which is always indicative of intravascular hemolysis.

Extravascular hemolysis: Physiological red cell destruction occurs in the liver or spleen so avoiding free hemoglobin in the plasma.

Causes of hemolytic anemia :

Inherited red cell abnormalities: resulting in chronic hemolytic anemia may arise from pathologies of the red cell membrane (hereditary spherocytosis or elliptocytosis).

Abnormal hemoglobin (hemoglobinopathies)
Abnormality in protective enzymes which prevent cellular oxidative damage, such as glucose-6-phosphate dehydrogenase (G6PD).
Acquired causes include auto- and allo-antibody-mediated destruction of red blood cells and other mechanical, toxic and infective causes.

Inherited red cell abnormalities:

Hereditary spherocytosis, hereditary elliptocytosis, hereditary ovalocytosis and hereditary stomatocytosis.

Red cell membrane defects:

cytoskeleton of RBC ensures great deformability and elasticity; the red cell diameter is 8 μm but the narrowest capillaries in the circulation are in the spleen, measuring just 2 μm in diameter. When the normal red cell structure is disturbed, usually by a quantitative or functional deficiency of one or more proteins in the cytoskeleton, cells lose their elasticity. Each time such cells pass through the spleen, they lose membrane relative to their cell volume. This results in an increase in mean cell hemoglobin concentration (MCHC), abnormal cell shape and reduced red cell survival due to extravascular hemolysis.

Hereditary spherocytosis:

Autosomal dominant condition, asymptomatic compensated chronic hemolytic state with spherocytes present on the blood film, a reticulocytosis and mild hyperbilirubinaemia. Pigment gallstones are present in up to 50% of patients and may cause symptomatic cholecystitis. Occasional cases are associated with more severe hemolysis.
H.S. shows marked heterogeneity, ranging from an asymptomatic condition to fulminant hemolytic anemia. the signs and symptoms of hereditary spherocytosis (HS) include mild pallor, intermittent jaundice, and splenomegaly is the common finding .

The clinical course may be complicated by crises:

- ***A hemolytic crisis*** occurs when the severity of hemolysis increases; this is rare, and usually associated with infection.
- ***A megaloblastic crisis*** follows the development of folate deficiency; this may occur as a first presentation of the disease in pregnancy.

- ***An aplastic crisis*** occurs in association with erythrovirus infection(Parvovirus B19)=**Fifth disease (erythema infectiosum)** .Erythrovirus causes a common exanthem in children, but if individuals with chronic hemolysis become infected, the virus directly invades red cell precursors and temporarily switches off red cell production. Patients present with severe anemia and a low reticulocyte count.

Investigations:

The blood film will show spherocytosis , MCHC is increased due to loss of membrane and the consequent spherical shape assumed by the cell.
direct Coombs test is negative excluding immune hemolysis ,
An osmotic fragility test show lysis in hypotonic saline solutions but is limited by lack of sensitivity and specificity.
More specific flow cytometric tests, detecting binding of eosin-5-maleimide to red cells, are recommended in borderline cases

Management: Folic acid prophylaxis, 5 mg once weekly, should be given for life**.** Splenectomy after age 7 years indications moderate to severe hemolysis with complications (anemia and gallstones),
Acute, severe hemolytic crises require blood transfusion, but blood must be cross-matched carefully and transfused slowly as hemolytic transfusion reactions may occur.

Hereditary Elliptocytosis: RBC membrane are characterized by elliptical-shaped erythrocytes.

The mode of inheritance is autosomal dominant. In most cases no symptoms and requires no therapy. For patients with clinically significant hemolytic anemia, splenectomy provides marked improvement.

Red cell enzymopathies:

production of energy via ATP. Anaerobic glycolysis via the Embden-Meyerhof pathway generates ATP hexose monophosphate shunt produces NADPH and glutathione to protect against oxidative stress.

Glucose-6-phosphate dehydrogenase (G6PD) deficiency:

This is usually inherited as an x linked inherited ,The deficiency therefore affects males and rare homozygotic females There are over 400 subtypes of G6PD described A+ A-, B+, B- The deficiency is more severe when enzyme levels 1% or less. This enzyme is important in the hexose monophosphate shunt pathway,

Clinical features: hemolytic anemias precipitating and presenting by the following

- Favism, i.e. acute hemolysis after ingestion of the fava bean
- Acute drug-induced hemolysis to (e.g.)
  - Analgesics: aspirin,paracetamol
  - Antimalarials: primaquine, quinine, chloroquine, pyrimethamine
  - Antibiotics: sulphonamides, nitrofurantoin, ciprofloxacin
  - Miscellaneous: quinidine, probenecid, vitamin K, dapsone
- Chronic compensated hemolysis
- Infection or acute illness
- Neonatal jaundice: may be a feature of the B^- enzyme

Investigations:

Non-spherocytic intravascular hemolysis during an attack

blood film will show

- Bite cells (red cells with a 'bite' of membrane missing)

- Blister cells (red cells with surface blistering of the membrane)
- Irregularly shaped small cells
- Polychromasia reflecting the reticulocytosis
- Denatured hemoglobin visible as Heinz bodies within the red cell cytoplasm, if stained with a supravital stain such as methyl violet.

G6PD level it is low in (G6PDD) Care must be taken close to an acute hemolytic episode because reticulocytes may have higher enzyme levels and give rise to a false normal result
Management aims to stop any precipitant drugs and treat any underlying infection. Acute blood transfusion may be life-saving.

Pyruvate kinase deficiency

This is the second most common red cell enzyme defect. It results in deficiency of ATP production and a chronic hemolytic anemia. It is inherited as an autosomal recessive trait. The extent of anemia is variable; the blood film shows characteristic 'prickle cells'. Enzyme activity is only 5-20% of normal. Transfusion support may be necessary.

Autoimmune hemolytic anemia

The antibodies may be IgG or M, or more rarely IgE or A. If an antibody fixes complement, it will cause intravascular hemolysis, but if complement activation is weak, the hemolysis will be extravascular(liver ,spleen) Antibody-coated red cells lose membrane to macrophages in the spleen and hence spherocytes are present in the blood.

- *Warm antibodies* bind best at 37 °C and account for 80% of cases. The majority are IgG
- *Cold antibodies* bind best at 4 °C but can bind up to 37 °C in some cases. They are usually IgM and bind complement. They account for the other 20% of cases.

Warm autoimmune hemolysis The majority are IgG, more common in middle age and in females. No underlying cause is identified in up to 50% of cases. The remainder are secondary to a wide variety of other condition
- Lymphoid neoplasms: lymphoma, chronic lymphocytic leukemia, myeloma
-Solid tumors: lung, colon, kidney, ovary, thymoma
-Connective tissue disease: SLE, rheumatoid arthritis -
-Drugs: methyldopa, mefenamic acid, penicillin, quinidine
-Miscellaneous: ulcerative colitis, HIV

Investigations: blood film evidence of hemolysis and spherocytosis**.** The diagnosis is confirmed by the direct Coombs(antiglobulin test)

Management:

If the hemolysis is secondary to an underlying cause, this must be treated and any offending drugs stopped.

Prednisolone 1 mg/kg orally. A response is seen in 70-80% of cases but may take up to 3 weeks; a rise in hemoglobin will be matched by a fall in bilirubin, LDH and reticulocyte levels. Once the hemoglobin has normalized and the reticulocytosis resolved, the corticosteroid dose can be reduced slowly over about 10 weeks. Corticosteroids work by decreasing macrophage destruction of antibody-coated red cells and reducing antibody production.

Blood Transfusion may be required for life-threatening problems, such as the development of heart failure

or rapid decrease in Hb.

Splenectomy should be considered If the hemolysis not respond to corticosteroids or can only be stabilised by large doses.

immunosuppressive therapy with azathioprine or cyclophosphamide may be considered

The anti-CD20 (B cell) monoclonal antibody, rituximab, has shown some success in difficult cases.

Cold agglutinin disease: is a rare form of autoimmune hemolytic anemia caused by cold-reacting autoantibodies**.** Antibodies, IgM, which bind to the red cells at 4 °C and cause them to agglutinate. It may cause intravascular hemolysis if complement fixation occurs. This can be chronic when the antibody is monoclonal, or acute or transient when the antibody is polyclonal.

Chronic cold agglutinin disease: This affects elderly patients and may be associated with an underlying low-grade B cell lymphoma. It causes a low-grade intravascular hemolysis with cold, painful and often blue fingers, toes, ears or nose (so-called acrocyanosis).

The blood film shows red cell agglutination and the MCV may be spuriously raised because the automated analyzers count aggregates as single cells.

The monoclonal IgM usually has specificity against the red cell antigen and is present in a very high titre

Treatment:

Treat underline disease and if idiopathic, then patients must keep extremities warm, especially in winter. Some patients respond to corticosteroid therapy and blood transfusion may be considered, but the cross-match sample must be placed in a transport flask at a temperature of 37 °C and blood administered via a blood-warmer. Other causes of cold agglutination can occur in association with Mycoplasma pneumoniae or with infectious mononucleosis

Alloimmune hemolytic anemia: unmatched transfusion of red cells (a hemolytic transfusion reaction. Maternal sensitization to paternal antigens on fetal cells (hemolytic disease of the newborn)**.**

Non-immune hemolytic anemia:

Physical trauma

- ***Mechanical heart valves***. High flow through incompetent valves or periprosthetic leaks through the suture ring holding a valve in place result in shear stress damage.
- ***March hemoglobinuria.*** Vigorous exercise, such as prolonged marching or marathon running, can cause red cell damage in the capillaries in the feet.
- ***Thermal injury***. Severe burns cause thermal damage to red cells characterized by fragmentation and the presence of microspherocytes in the blood.
- ***Microangiopathic hemolytic anemia***. Fibrin deposition in capillaries can cause severe red cell disruption. It may occur in a wide variety of conditions: disseminated carcinomatosis, malignant or pregnancy-induced hypertension, hemolytic uremic syndrome ,thrombotic thrombocytopenic purpura ,and disseminated intravascular coagulation

Infection: Plasmodium falciparum malaria ,may be associated with intravascular hemolysis; when severe, this is termed black water fever due to the associated hemoglobinuria. Clostridium perfringens septicemia

Chemicals or drugs: Dapsone and sulfasalazine cause hemolysis by oxidative denaturation of hemoglobin. Denatured hemoglobin forms Heinz bodies in the red cells, visible on supravital staining with

brilliant cresyl blue. Arsenic gas, copper, chlorates, nitrites and nitrobenzene derivatives may all cause hemolysis.

Paroxysmal Nocturnal Hemoglobinuria (PNH)

It is a rare, acquired, potentially life-threatening disease of the blood characterized by complement-induced intravascular hemolytic anemia ,red-brown urine (due to the appearance of hemoglobin in the urine) The disease is associated with an increased risk of venous thrombosis as pul. embolism and unusual sites such as the liver or abdomen. CNS. PNH is also associated with hypoplastic bone marrow failure, aplastic anemia and myelodysplastic syndrome PNH is the only hemolytic anemia caused by an acquired (rather than inherited) intrinsic defect in the cell membrane (deficiency of glycophosphatidylinositol leading to absence of protective proteins on the membrane).

Investigation

Intravascular hemolysis: low Hb, high LDH, high retic count, high bilirubin(indirect),low haptoglobin,-ve coombs test, microcytic RBC

Hams test The test involves placing red blood cells in mild acid; a positive result (increased RBC fragility)

Confirmation by flow cytometry for CD55 and CD59 on WBC and RBC

Management: It is supportive with Folic acid supplements ,blood transfusion and treatment of thrombosis. Recently the anti-complement C5 monoclonal antibody ecluzimab(Soliris) was shown to be effective in reducing haemolysis.and reducing the need for blood transfusions, improving quality of life, and reducing the risk of thrombosis**.**

is the only curative therapy,Allogeneic bone marrow transplantation