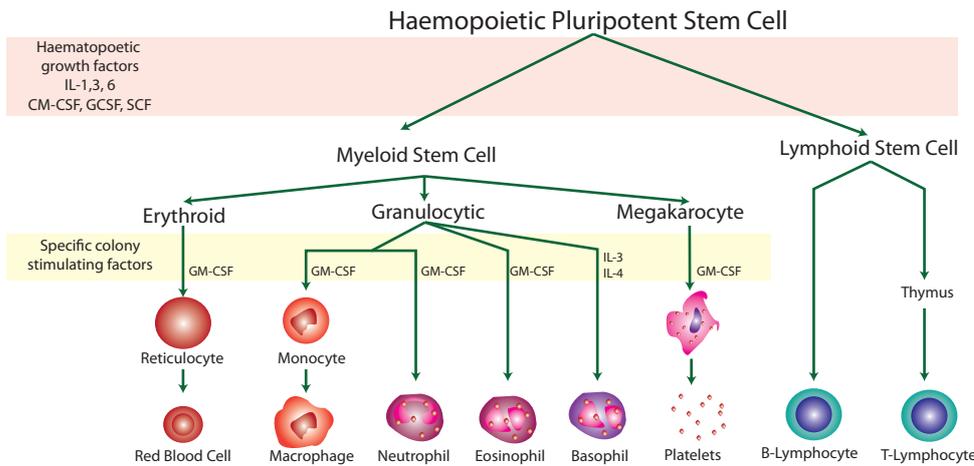


| | |
|-------------|-----|
| Na+ | 142 |
| K+ | 4.2 |
| Ca2+ | 1.3 |
| Mg2+ | 0.8 |
| Cl- | 108 |
| HCO3- | 24 |
| phosphates | 2 |
| SO4- | 0.5 |
| amino acids | 2 |
| creatine | 0.2 |
| lactate | 1.2 |
| glucose | 5.6 |
| protein | 1.2 |
| urea | 4 |
| other ions | 4.8 |

Plasma constituents comprises about 18% of extracellular fluid, or 5% of bodyweight. It is the noncellular part of the blood, being about 60% of blood volume. It is 0.7% solids, so measured concentrations of ions per litre plasma are lower than the actual concentrations per litre of water present. The ionic composition of plasma is similar to that of interstitial fluid, except for a higher protein concentration and (because of the Gibbs-Donnan equilibrium) a slightly higher concentration of diffusible cations. Typical constituents (in mOsm/l) are shown adjacent. The osmolality of protein in plasma is low but the high molecular weight of proteins means that they are a major constituent of plasma when measured by weight. The protein constituent of plasma can be fractionated by electrophoresis into albumin and α , β and γ globulins and other proteins such as fibrinogen. Albumin makes up over half the protein content by weight and 75% of the oncotic pressure. It and a number of globulin proteins bind circulating hormones and drugs. Albumin also provides a source of aminoacids to tissues. The globulin proteins include many specialized binding proteins, the immunoglobulins and enzymes such as the clotting factors. Fibrinogen plays an important role in blood clotting.

The cellular elements of blood are derived from the bone marrow and lymphoid tissue (and liver and spleen in the neonate). Putative pluripotent stem cells in the marrow give rise to lymphoid and myeloid stem cells. Lymphoid stem cells differentiate into pro-T and pro-B lymphoid cells which mature in the thymus or lymph nodes into T and B lymphocytes (and plasma cells). Lymphoid cell lines are distinguishable only by cell surface markers. Myeloid stem cells differentiate into colony forming units (CFU) of the erythroid, granulocytic and megakaryocyte lines and the influx of colony stimulating factors. The myeloid and erythroid cell lines differentiate through several morphologically distinct stages. The erythroid line begins with the proerythroblast, a nucleated cell devoid of haemoglobin which gradually increases its haemoglobin content and reduces its size, finally losing all cytoplasmic organelles as a reticulocyte. The differentiation from stem cell to erythrocyte takes about a week. Haemoglobin is synthesized in erythroid cells from the proerythroblast stage. It consists of four globin chains ($\alpha_2\beta_2$ in adult Hb) each covalently linked to a haem molecule. Haem is synthesized from glycine, succinyl-SCoA and Fe²⁺. Synthesis is commonly limited by Fe deficiency, resulting in hypochromic microcytic anaemia. The rate-limiting step in haem synthesis is the condensation of succinyl-SCoA and glycine to form δ -aminolaevulinic acid. This step also requires pyridoxine. 2 δ -aminolaevulinic acid molecules are condensed to form a pyrrole ring, 4 of

which are required to form protoporphyrin IX. This coordinates with four of the six coordination points on Fe²⁺, to form haem. The globin chains are synthesized in the RER of erythroid cells, each having a molecular weight of about 16,000. 97% of haemoglobin in normal adults is Hb A ($\alpha_2\beta_2$), most of the remaining 3% is Hb A2 ($\alpha_2\delta_2$), with very small amounts of Hb F ($\alpha_2\gamma_2$). These proportions are different in thalassaemia, where the production of one chain type is disordered. Plasma proteins can be subdivided by electrophoresis into α_1 , α_2 , β_1 , β_2 and γ fractions and fibrinogen. All the fractions are produced almost entirely in the liver, with the exception of γ -globulins which are produced in lymph nodes and small quantities of other proteins and peptide hormones which are produced in many different organs. Albumin turnover is about 200-400 mg/kg/day from a total pool of 4-5 g/kg.



Abnormal haemoglobin There are five major classes of hemoglobinopathies. Structural hemoglobinopathies occur when mutations alter the amino acid sequence of a globin chain, altering the physiologic properties of the variant hemoglobins and producing the characteristic clinical abnormalities. The most clinically relevant variant are the sickle cell syndromes which are caused by a mutation in the β -globin gene that changes the sixth amino acid from glutamic acid to valine. The prototype disease, sickle cell anemia, is the homozygous state for HbS and clinically results in persistent anaemia due to a red cell survival time of only 10-15 days. They suffer multiple painful infarcts, particularly to the renal medulla and spleen, and may have aplastic crises precipitated by folate deficiency or by infection. Hb S has a reduced affinity for oxygen, so homozygotes have reduced saturation for PO₂ and display increased physiological shunt. Crises are precipitated by hypoxia, hypovolaemia and cold. Hb C and Hb M are other structural hemoglobinopathies. There are also Hb variants which demonstrate either high or low affinity to O₂. The former may lead to reduced tissue O₂ delivery but the latter is usually asymptomatic. Thalassaemia syndromes are inherited disorders of α - or β -globin biosynthesis. The reduced supply of globin diminishes production of hemoglobin tetramers, causing hypochromia and microcytosis. Unbalanced accumulation of α and β subunits occurs because the synthesis of the unaffected globins proceeds at a normal rate. Unbalanced chain accumulation dominates the clinical phenotype. Clinical severity varies widely, depending on the degree to which the synthesis of the affected globin is impaired, altered synthesis of other globin chains, and co-inheritance of other abnormal globin alleles. Clinical manifestations range from asymptomatic anemia, microcytosis and hypochromia through to severe debilitating anaemia and non compatibility with life in the case of hydrops fetalis. Thalassaemic hemoglobin variants combine features of thalassaemia (e.g., abnormal globin biosynthesis) and of structural hemoglobinopathies (e.g., an abnormal amino acid sequence). Hereditary persistence of fetal hemoglobin (HPFH) is characterized by synthesis of high levels of fetal hemoglobin in adult life. Acquired hemoglobinopathies include modifications of the hemoglobin molecule by toxins (e.g., acquired methemoglobinemia) and abnormal hemoglobin synthesis (e.g., high levels of HbF production in preleukemia and α thalassaemia in myeloproliferative disorders).

Haemoglobinopathies Classification

1. Structural hemoglobinopathies
Sickle cell anaemias
Hb C and M
Low and high O₂ affinity Hb
2. Thalassaemias
Alpha thalassaemia variants
Beta thalassaemia variants
3. Combined structural/thalassaemias
4. Hereditary persistence of fetal Hb (HPFH)
5. Acquired Haemoglobinopathies
Methemoglobinemia
Leukaemia induced disorders of Hb

Assessing platelet function, coagulation and fibrinolysis. A functional test of clotting is a bleeding time. A standardized cut is made on the skin and the time of bleeding measured. Unfortunately this is a difficult test to calibrate. It is a good test of platelet function as the formation of a platelet plug is usually the reaction limiting the duration of bleeding, but if the time is prolonged, it does not indicate the nature of the clotting defect. A platelet count has good predictive value of the risk of bleeding if the platelets are known to be of normal function. A count below $50 \times 10^9 / l$ is associated with substantially prolonged bleeding time and below $20 \times 10^9 / l$ is associated with dangerous spontaneous haemorrhages. The function of the extrinsic and common pathways is assessed by prothrombin time (or INR). A specimen of plasma at 37° is citrated (to bind any ionized Ca²⁺) and an excess of tissue factor and Ca²⁺ are added as a timer is started. The time taken to coagulate is an most commonly used to assess the effect of coumarin anticoagulants which impair synthesis of factors VII, IX, X and prothrombin. The function of the intrinsic and common pathways is commonly measured using activated partial thromboplastin time (APTT). Citrated plasma at 37° is combined with kaolin and cephalin before an excess of Ca²⁺ is added and the time taken to coagulate is measured. This screens for the adequacy of factors XII, XI, IX, VII, PK and HMWK as well as the common pathway. It is used to adjust the dose of heparin used for anticoagulation. The common pathway is assessed using a thrombin time (TT). Thrombin as added to plasma and the time taken to coagulate measured. Ca²⁺ is not required for coagulation in this test. In cardiopulmonary bypass, large doses of heparin are used, and clotting is assessed using the activated clotting time (ACT), an automated device optimized for assessing supratherapeutic heparinization. ACT is in excess of 400 s for bypass. None of these tests will detect factor XIII or alpha2-antiplasmin deficiency and they may miss vWF deficiency which can be cyclical, so they cannot exclude the possibility of excessive bleeding. There is also a substantial functional reserve in the concentration of most clotting factors. In haemophilia A, symptoms are uncommon while the factor VIII level is above 5% of normal. To determine a specific cause for defective clotting, it is necessary to do specific factor assays and test for the presence of anticoagulant factors such as anti-factor antibodies. The function of the fibrinolytic system can be assessed by clot lysis time. This is shortened in alpha2-antiplasmin deficiency. Circulating fibrin degradation products can be assayed and give some information about the amount of clot lysis occurring. Fibrin crosslinking can be assessed by clot solubility in 5 M urea, which is increased in factor XIII deficiency.