Blood

Introduction: Blood has three main functions

1) transportation
2) regulation
3) protection

Transportation: Blood is responsible for the transport of oxygen from the lungs to the tissues and carbon dioxide from the tissues to the lungs. Blood also carries nutrients from the gastrointestinal tract to the cells and carries cellular waste products away from the cells. Blood is also responsible for transporting hormones (which are produced in endocrine glands) to the cells. Another function of blood, which is often overlooked, is heat transfer.

Regulation: Blood helps maintain the required pH of body tissues through its buffering capacity. Blood helps regulate temperature through its heating and cooling capacities. It also regulates the water content in cells mainly through dissolved sodium ions and proteins.

Protection: Elements in the blood help to prevent blood loss by clotting. Blood also helps to protect against foreign microbes through the action of white blood cells and special blood proteins such as antibodies, interferon, and complement.

Physical properties of blood: Blood is heavier and thicker than water, it therefore flows slower than water. Blood has a viscosity of 4.5 to 5.5 versus a viscosity of 1 for water. Blood also maintains a temperature slightly higher than that of the body (38°C or 100.4°F). It maintains a slightly alkaline pH (7.35 to 7.45) and a salt concentration of 0.9%.

Starting with the whole body: blood makes up about 8% of the entire body weight. (5-6 liters for the average male and 4 to 5 liters for the average female).

Let us first talk about plasma, and get it out of the way.

Plasma is the straw colored liquid which remains when we remove the formed elements from the blood. Plasma is NOT the same as serum. Serum is plasma which has had the clotting elements removed.

Plasma is 91.5% water and 8.5% solutes (proteins). Many of the proteins found in plasma are also found elsewhere in the body, but when found in the plasma they are referred to as plasma proteins.

Plasma Proteins

1) Albumin: 55%, is synthesized in the liver, Concentration in the plasma is 4X higher than in interstitial fluids. This is mainly what gives blood its viscosity. Albumin helps to maintain the osmotic balance of the body by allowing excess tissue fluid to move into the blood or fluid to move from the blood into the tissues. Water moves from areas of high water concentration (low solute) to areas of low water concentration (high solute).

2) Globulins: 38%, are synthesized in the liver and in plasma cells. Mainly belong to the group of proteins called immunoglobulins (antibodies). Will learn more about this when you cover the immune system.

3) Fibrinogen: 7%, produced in the liver, it plays a key role in clotting mechanisms.

Other Solutes in Plasma

1) NPN: (non protein nitrogens). These contain nitrogen but they are not proteins. Mainly these are metabolic wastes such as urea, uric acid, creatine, creatinine, ammonium salts, bilirubin.
2) Nutrients: the products of digestion which are to be delivered to the tissues. Includes Amino acids (from protein breakdown), glucose (from carbohydrates), and fatty acids and glycerol (from triglycerides).

3) Regulatory substances: Enzymes to help catalyze chemical reactions (each cell is a miniature laboratory), and hormones to regulate growth and development.

4) Respiratory gases: although oxygen is related to the hemoglobin in the RBCs, carbon dioxide is more closely associated with plasma in which it is dissolved (Coke Shuttle Experiments).

5) Electrolytes: these are inorganic salts such as Na+, K+, Ca+, Mg+ (cations) and Cl-, HPO43-, SO42-, HCO_3- (anions). These help to maintain the proper osmotic pressure, normal pH, and physiological balance between the blood and tissues.

The Formed Elements

Erythrocytes (RBCs): 99% of the formed elements. Are biconcave discs of about 8 µm diameter. These cells lack a nucleus and organelles. They cannot reproduce or carry on extensive metabolic activity. The biconcave shape greatly increases the surface area of the cell and also allows flexibility of the cell, which may pass through capillaries no wider than 3 µm in diameter. A healthy male has approximately 5.4 million RBCs/mm3 of blood. A healthy female has approximately 4.8 million RBCs/mm3 of blood. The higher number of RBCs in males is due to the presence of testosterone, which stimulates the production of RBCs through its stimulation of the synthesis of erythropoietin.

The cell membrane of the RBC has certain markers (antigens) that are responsible for the various blood groups.

RBCs contain hemoglobin which accounts for about 33% of the weight of an RBC and each RBC has about 280 million hemoglobin molecules. Since RBCs are non-nucleated all of their internal space is available for oxygen transport. Also, since they contain no mitochondria and generate ATP anaerobically (without oxygen) they do not consume any of the oxygen that they carry. Normal values for hemoglobin are about 14 - 20 g/100 ml blood in infants, 12 - 15 g/100 ml in adult females, and 14 - 16.5 g/100 ml in adult males.

Hemoglobin combines with oxygen to form oxyhemoglobin and with carbon dioxide to form carbaminohemoglobin. Each hemoglobin molecule consists of 4 heme groups and 1 globin (protein). Each heme group can reversibly bind with oxygen and the globin reversibly binds with carbon dioxide.

RBCs have a life span of 120 days, after which they are removed from circulation by fixed macrophages in the spleen and liver. To maintain normal levels of RBCs the body must produce them at the rate of approximately 2 million per second.

Leukocytes: WBCs

Granular Leukocytes:

Neutrophils: 10 - 12 µm diam. Nuclei have 2 - 6 lobes connected by thin strands. As a neutrophil ages its nucleus becomes more lobulated. This gives us the name polymorphonuclear leukocytes or (PMNs). The cytoplasm contains many small, pale lilac colored granules.

Eosinophils: 10 - 12 µm diam. Nuclei have two lobes connected by a thick strand. Cytoplasm contains many large red-orange colored granules. These granules DO NOT obscure the nucleus.

Basophils: 8 - 10 µm diam. Nucleus is bilobed (irregular) and often S shaped. Granules in the cytoplasm stain blue-black, are variable in size, and usually obscure the nucleus.

Agranular: no cytoplasmic granules can be seen under the light microscope. Very little cytoplasm in comparison to nucleus.

Lymphocytes: 7 - 15 µm diam. Nuclei are darkly stained, usually round with a slight indentation. Cytoplasm appears as a thin sky blue rim around the nucleus.

T-cells: thymus activated

B-cells: bursa derived

Monocytes: 14 - 19 µm diam. Nuclei are usually kidney shaped. Cytoplasm appears as a foamy blue area. Will migrate out of the blood and become macrophages.

WBC Physiology

The primary function of WBCs is to combat infection from invading pathogens. This is usually accomplished through phagocytosis or immune responses. The two cells responsible for the majority of phagocytosis are neutrophils and macrophages and these cells are sometimes referred to as phagocytes. Phagocytes are drawn toward an area of infection by a process known as chemotaxis. This is a process in which chemicals from the inflamed tissue signal phagocytes to move into the area. In order for leukocytes to carry out their functions most must leave the blood stream. This is accomplished by a process known as diapedesis. Briefly, this involves passing through the capillary wall using a cytoplasmic flow type of motion.

Neutrophils respond to bacterial infection faster than the other WBC types. After phagocytosis the neutrophil will release several destructive chemicals such as lysozyme (anti-bacterial), and anti-oxidants such as superoxide anion (O_2-), hydrogen peroxide (H_2O_2) and hypochloride anion (OCl-). Neutrophils also contain defensins which are essentially spears which poke holes in the membranes of invading cells.

Monocytes take longer to reach the site of an infection, but they ultimately arrive in more numbers and can destroy more microbes. Monocytes themselves do little to combat infection, however upon reaching the site of the infection they differentiate and become macrophages. Macrophages clean up cellular debris and microbes.

Eosinophils also leave the capillaries and enter the tissue fluid. These release histaminase which counteracts the effects of histamine. Eosinophils also phagocytize antigen-antibody complexes. Will learn more when covering the immune system.

Basophils Leave the capillaries and develop into mast cells. Mast cells liberate heparin, histamine, and seratonin. These substances intensify the inflammatory reaction.

T and B cells are the main players in the immune system. You will learn more details about the immune system later. Briefly, any substance that stimulates an immune response is called an antigen. In response to an antigen B cells will develop into plasma cells which will produce antibodies. Antibodies bind with antigens in such a way that their binding sites are no longer available for other chemical reactions, thus rendering them inert. This is called an antigen-antibody complex. Antigens also stimulate T cells some of which (cytotoxic "killer" T cells) can destroy invaders directly. Others (T helper cells) assist both B and cytotoxic T cells.

Although B and T cells are our main defense against foreign invaders, they are also responsible for transfusion rejections, allergies, and organ transplant rejection. Due to their function of fighting infection most WBCs have a variable life span. Some may live for years in a very healthy individual, but the usual life span is only a few days. In the case of infection the life span may be shortened to only a few hours.

Thrombocytes: (platelets) 2 - 4 µm diam. Are not actually cells, but are pieces of cells. They are fragments of a very large cell found in the bone marrow, the megakaryocyte. Platelets carry chemicals which help promote blood clotting. The normal life span of a platelet is 5 - 9 days.

Now that we know what we have in the blood. Let us review where these constituents come from.

Hematopoiesis: (hemopoiesis) is the term given to the formation of blood cells. During fetal life hemopoiesis takes place in the yolk sac, liver, spleen, thymus gland, lymph nodes, and bone marrow. After birth hemopoiesis takes place in the red bone marrow. This marrow is found in the proximal epiphyses of the humerus and femur, the flat bones of the sternum, ribs, and cranium, and the vertebrae and pelvis.

All of the formed elements originate from a single cell type, the pluripotent hematopoietic stem cell. There are several hematopoietic growth factors which stimulate differentiation along certain pathways and therefore proliferation of certain progenitor cells.

> Erythropoietin: produced mainly in the kidneys and to some extent in the liver, stimulates proliferation of erythrocyte precursors.
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> Thrombopoietin: stimulates the formation of platelets.
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> Cytokines: A variety of glycoproteins which are produced by red bone marrow cells, leukocytes, macrophages, and fibroblasts, to name a few. The most famous of the cytokines are colony stimulating factors and interleukins.

Hemostasis: refers to the stoppage of bleeding. There are three mechanisms involved in hemostasis.

> 1) Vascular spasm: Caused by the contraction of the smooth muscles in the wall of the damaged vessel. This limits the amount of blood flow in the vessel.
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> 2) Platelet plug formation: Can be divided into three phases:
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> > a) *Platelet adhesion*: Platelets stick to parts of the damaged blood vessel such as the collagen underneath the endothelial cells.
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> > b) *Platelet release action*: Release of the contents of platelet granules in response to platelet adhesion. The platelets extend many projections which enable them to contact each other. Platelet granules contain substances such as clotting factors, Platelet derived

growth factor, ADP, ATP, Ca 2+, seratonin, thromboxane A2, and fibrin stabilizing factor to name a few.

c) *Platelet aggregation*: The release of ADP makes other platelets in the area sticky, and they stick to the originally activated platelets. Eventually this accumulation of platelets forms a mass called a platelet plug.

3) Blood coagulation (clotting) A blood clot is a network of fibrin fibers in which the formed elements are trapped. Clotting is a very complex process in which coagulation factors activate each other in a cascade. Basically clotting can be broken down into three stages.

a) formation of prothrombinase
b) conversion of prothrombin
c) conversion of fibrinogen

Although vitamin K itself is not involved in clot formation, it is required for the synthesis of 4 clotting factors. Therefore a Vitamin K deficiency may result in uncontrolled bleeding.

Fibrinolysis is the dissolution of a clot, this works through an enzyme built into the clot called plasminogen.

Clinical Correlations

Blood groups: Blood groups are determined by genetically determined agglutinogens (or isoantigens) found on the surface of the RBCs. There are at least 14 blood group systems, the most well known being the ABO group and the Rh group.

ABO groups: Based on the agglutinogens A and B. People who have agglutinogen A are type A, those with agglutinogen B are type B, those with agglutinogens A and B are type AB, and those who lack agglutinogens are type O. Genetically each person inherits one agglutinogen gene from each parent so there are six possible agglutinogen combinations (OO, AO, AA, BO, BB, and AB). A and B are dominant traits whereas O is recessive so that leaves us with four possible ABO groups.

OO = type O
AO and AA = type A
BO and BB = type B
AB = type AB.

The plasma of people who are A, B, or AB contains naturally occurring antibodies called agglutinins (or isoantibodies). These will react with A or B agglutinogens if the two are mixed. When these antigen-antibody complexes form in the blood they activate a system known as the complement fixation pathway. The end result of complement fixation is the formation of holes in the target cell (in this case the donated RBCs) which causes them to burst and release their hemoglobin into the plasma. This release of hemoglobin is referred to as hemolysis, and often results in kidney damage.

Agglutination: the clumping that occurs when incompatible blood types are mixed outside of the body. This is not hemolysis and it is not same as clotting.

Rh blood groups: are so named because they were first described in Rhesis monkeys. Like the ABO group it is based on antigens found on the surface of RBCs. People who have Rh agglutinogens (D antigens) are Rh+. Those without are Rh-. This system is different in that, normally plasma does not contain anti-Rh agglutinins. If an Rh- person receives Rh+ blood the body will start to make anti-Rh

agglutinins which will remain in the blood. If a second transfusion of Rh+ blood is given then hemolysis will occur.

Hemolytic Disease of the Newborn: (erythroblastosis fetalis) This is the most common problem associated with Rh incompatibility. It is only a concern if the mother is Rh-. Normally there is no exchange of blood across the placenta, however if Rh+ blood from a fetus leaks across into the bloodstream of the mother she will start to make anti-Rh agglutinins. Normally the baby of this pregnancy is unaffected, however if the mother becomes pregnant again her anti-Rh agglutinins can leak across the placenta and end up in the bloodstream of the fetus. If the fetus is Rh- there will be no problem since Rh- blood does not have the Rh agglutinogen. If the fetus is Rh+ hemolysis may occur. This condition is now avoided by giving all Rh- mothers an injection of anti-Rh gamma globulin (RhoGAM) shortly after birth. This binds to the fetal agglutinogens if present, so that the mother's immune system will not respond to them. This protects the fetus of the next pregnancy.

Clinical Correlations

Erythrocytes:

Hematocrit: The percentage or RBCs in blood. Normally used to diagnose anemia and polycythemia (abnormally high % RBCs) and abnormal states of dehydration. Females = 38 - 46%, males 40 - 54%. Low hematocrit may signal anemia. High >65% signals polcythemia. Athletes and people who live in high mountain regions usually have a higher than average hematocrit.

Reticulocyte count: a measurement of the rate of erythropoiesis determined by counting the % of reticulocytes present in the blood.

Blood doping: The practice of removing and storing RBCs to be reinjected prior to an athletic event. This results in increased oxygen delivery to the muscles. This practice causes increased viscosity of the blood which causes the heart to become over worked.

WBCs:

Differential White Cell Count: A count of the various WBCs present in the blood. This can be compared to the normal values for WBCs. A rise or fall in the percentages of the various cells can be indicative of various conditions.

Thrombocytes:

Hemophilia: A name given to several different hereditary deficiencies in coagulation. Basically one or more factors of the coagulation pathway are deficient so clotting does not occur. Treatment includes transfusion of fresh plasma or concentrates of the deficient factors. Contaminated (HIV) blood (1982-1985) given to hemophiliacs resulted in those people becoming infected with HIV.

Streptokinase: A clot dissolving agent given to patient to restore circulation in blocked arteries.

Tissue plasminogen activator: a genetically engineered version of streptokinase. Very expensive, but not any better.

Clinical Terms/Conditions

Anemia – is a condition in which the oxygen carrying capacity of the blood is reduced. All forms of anemia have either reduced RBC numbers or reduced levels of hemoglobin.

Nutritional anemia – is due to an inadequate diet, i.e., not enough iron, amino acids, Vitamin B12

Pernicious anemia – the patient is lacking intrinsic factor and therefore cannot absorb Vitamin B12

Hemorrhagic anemia – is due to a loss of blood (bleeding)

Hemolytic anemia – is due to the rupture of RBC's. Hemoglobin spills into the blood

Thalassemia – is a form of hemolytic anemia. Here we see defects in hemoglobin synthesis and thin, fragile RBCs. The condition is more common in Mediterranean areas.

Aplastic anemia – here we see low levels of RBCs related to red bone marrow that is being replaced by fatty tissue, fibrous tissue, or tumor cells

Sickle-Cell anemia – Here we see irregular shaped RBCs. Low oxygenation causes the sickle shape. These irregular shaped RBC lodge in smaller vessels. They also easily rupture thus causing hemolytic anemia. Potassium easily leaks from these cells, which provides an interesting advantage to this form of anemia, as potassium kills malarial parasites.

Polycythemia – is an elevated RBC count. It increases blood viscosity which can lead to high blood pressure and hemorrhage.

Mononucleosis (infectious) – is caused by the Epstein-Barr virus. The virus is transmitted orally, which give mononucleosis the nick-name "the kissing disease." The virus multiplies in lymphatic tissues (B cells) and causes the B cells to enlarge to the size of monocytes. The accumulation of monocytes in the spleen can lead to splenic rupture, so people with this condition usually have to limit their level of physical activity until the condition passes.

Leukemia – there are two initial forms of leukemia, acute and chronic.

Acute – this is an uncontrolled production and accumulation of immature leukocytes

Chronic – this an accumulation of mature leukocytes. In this condition the leukocytes have outlived their normal life span