PHLEBOTOMY INTRO

The Vascular System
The vascular system is the system of blood vessels that, along with the heart, forms the closed loop through which blood is circulated to all parts of the body. There are two divisions to this system, the pulmonary circulation and the systemic circulation.

The Pulmonary Circulation
The pulmonary circulation carries blood from the right ventricle of the heart to the lungs to remove carbon dioxide and pick up oxygen; the oxygenated blood is then returned to the left atrium of the heart.

The Systemic Circulation
The systemic circulation serves the rest of the body, carrying oxygenated blood and nutrients from the left ventricle of the heart to the body cells and then returning to the right atrium of the heart with blood carrying carbon dioxide and other waste products of metabolism from the cells.

STRUCTURES
The structures of the vascular system are the various blood vessels that, along with the heart, form the closed system through which blood flows. Blood vessels are tube-like structures capable of expanding and contracting. According to information from the Arizona Science Center, the human vascular system has around 250,000 miles of blood vessels, 95% of which are capillaries, which make up what is called the capillary bed. The rest are arteries and veins.

Arteries
Arteries are blood vessels that carry blood away from the heart. They have thick walls because the blood that moves through them is under pressure from the contraction of the ventricles. This pressure creates a pulse that can be felt, distinguishing the arteries from the veins.

**KEY POINT** When arterial blood is collected by syringe, the pressure normally causes the blood to “pump” or pulse into the syringe under its own power.

**KEY POINT** The pulmonary artery is the only artery that carries deoxygenated, or oxygen-poor, blood. It is part of the pulmonary circulation and carries deoxygenated blood from the heart to the lungs. It is classified as an artery because it carries blood away from the heart.

The smallest branches of arteries that join with the capillaries are called arterioles (ar-te’-re- olz). The largest artery in the body is the aorta. It is approximately 1 inch (2.5 cm) in diameter.

Veins
Veins are blood vessels that return blood to the heart. Veins carry blood that is low in oxygen (deoxygenated or oxygen-poor) except for the pulmonary vein, which carries oxygenated blood from the lungs back to the heart. Because systemic venous blood is oxygen-poor, it is much darker and more bluish-red than normal arterial blood.

The walls of veins are thinner than those of arteries because the blood is under less pressure than arterial blood. Since the walls are thinner, veins can collapse more easily than arteries. Blood is kept moving through veins by skeletal muscle movement, valves that prevent the backflow of blood, and pressure changes in the abdominal and thoracic cavities during breathing.

The smallest veins at the junction of the capillaries are called venules (ven’ulz). The largest veins in the body are the **venae cavae** (singular, vena cava). The longest veins in the body are the **great saphenous** (sa-fe’nus) veins in the leg.

Capillaries
Capillaries are microscopic, one-cell-thick vessels that connect the arterioles and venules, forming a bridge between the arterial and venous circulation. Blood in the capillaries is a mixture of both venous and arterial blood. In the systemic circulation, arterial blood delivers oxygen and nutrients to the capillaries. The thin capillary walls allow the exchange of oxygen for carbon dioxide and nutrients for wastes between the cells and the blood. Carbon dioxide and wastes are carried away in the venous blood. In the pulmonary circulation, carbon dioxide is delivered to the capillaries in the lungs and exchanged for oxygen.

**BLOOD VESSEL STRUCTURE**
Arteries and veins are composed of three main layers. The thickness of the layers varies with the size and type of blood vessel. Capillaries are composed of a single layer of endothelial cells enclosed in a basement membrane.

Layers
• **Tunica (tu’ni-ka) adventitia** (ad’ven-tish’e-a): the outer layer of a blood vessel, some times called the tunica externa. It is made up of connective tissue and is thicker in arteries than in veins.
• **Tunica media**: the middle layer of a blood vessel. It is made up of smooth muscle tissue and some elastic fibers. It is much thicker in arteries than in veins.

• **Tunica intima** (in’ti-ma): the inner layer or lining of a blood vessel, sometimes called the tunica interna. It is made up of a single layer of endothelial cells with an underlying basement membrane, a connective tissue layer, and an elastic internal membrane.

Valves

Venous valves are thin membranous leaflets composed primarily of epithelium similar to that of the semilunar valves of the heart. Most of the venous system flows against the pull of gravity. As blood is moved forward by the movement of skeletal muscle, for example, the valves help keep it flowing toward the heart by allowing blood to flow in only one direction.

---

**KEY POINT** The presence of valves within veins is a major structural difference between arteries and veins.

**PHLEBOTOMY-RELATED VASCULAR ANATOMY**

**Antecubital Fossa**

Antecubital (an’te-ku’bi-tal) means “in front of the elbow.” **Fossa** means a shallow depression. The **antecubital** (AC) **fossa** is the shallow depression in the arm that is anterior to (in front of) and below the bend of the elbow. It is the first-choice location for venipuncture because several major arm veins lie close to the surface in this area, making them relatively easy to locate and penetrate with a needle. These major superficial veins are referred to as **antecubital veins**. The anatomical arrangement of antecubital veins varies slightly from person to person; however, two basic vein arrangements, referred to as the H- and M-shaped patterns are seen most often.

**H-Shaped Antecubital Veins**

The H-shaped venous distribution pattern is displayed by approximately 70% of the population and includes the median cubital vein, cephalic vein, and basilic vein.

• **Median cubital vein**: Located near the center of the antecubital area, it is the preferred vein for venipuncture in the H-shaped pattern. It is typically larger, closer to the surface, better anchored, and more stationary than the others, making it the easiest and least painful to puncture and the least likely to bruise.

• **Cephalic vein**: Located in the lateral aspect of the antecubital area, it is the second-choice vein for venipuncture in the H-shaped pattern. It is often harder to palpate than the median cubital but is fairly well anchored and often the only vein that can be palpated (felt) in obese patients.

• **Basilic vein**: A large vein located on the medial aspect (inner side) of the antecubital area, it is the last-choice vein for venipuncture in either venous distribution pattern. It is generally easy to palpate but is not as well anchored and rolls more easily, increasing the possibility of accidental puncture of the anterior or posterior branch of the **medial cutaneous nerve** (a major nerve of the arm) or the **brachial artery**, both of which commonly underlie this area. Punctures in this area also tend to be more painful.

**M-Shaped Antecubital Veins**

The veins that form the M-shaped venous distribution pattern include the cephalic vein, median vein, median cephalic vein, median basilic vein, and basilic vein. The veins most commonly used for venipuncture in this distribution pattern are described as follows:

• **Median vein** (also called the **intermediate antebrachial vein**): The first choice for venipuncture in the M-shaped pattern because it is well anchored, tends to be less painful to puncture, and is not as close to major nerves or arteries as the others, making it generally the safest one to use.

• **Median cephalic vein** (also called the **intermediate cephalic vein**): The second choice for venipuncture in the M-shaped pattern because it is accessible and is for the most part located away from major nerves or arteries, making it generally safe to puncture. It is also less likely to roll and relatively less painful to puncture.

• **Median basilic vein** (also called the **intermediate basilic vein**): The last choice for venipuncture in the M-shaped pattern (even though it may appear more accessible) because it is more painful to puncture and, like the basilic vein, is located near the anterior and posterior branches of the median cutaneous nerve and the brachial artery.

---

**KEY POINT** Vein location differs somewhat from person to person, and you may not see the exact textbook pattern. The important thing to remember is to choose a prominent vein that is well fixed and does not overlie a pulse, which indicates the presence of an artery and the potential presence of a major nerve.

**Other Arm and Hand Veins**

According to CLSI, although the larger and fuller median, median cubital, cephalic, and median cephalic veins are used most frequently, veins on the back of the hand and wrist are also acceptable for venipuncture.

---

**CAUTION**: Veins on the underside of the wrist are never acceptable for venipuncture.
Leg, Ankle, and Foot Veins
Because of the potential for significant medical complications such as phlebitis or thrombosis, veins of the leg, ankle, and foot must not be used for venipuncture without permission from the patient’s physician. Puncture of the femoral vein is performed only by physicians or specially trained personnel.

Arteries
Arteries are not used for routine blood collection. Arterial puncture requires special training to perform, is more painful and hazardous to the patient, and is generally limited to the collection of arterial blood gas (ABG) specimens for the evaluation of respiratory function.

THE BLOOD
Blood has been referred to as “the river of life,” as it flows throughout the circulatory system delivering nutrients, oxygen, and other substances to the cells and transporting waste products away from the cells for elimination.

BLOOD COMPOSITION
Blood is a mixture of fluid and cells; it is about five times thicker than water, salty to the taste, and slightly alkaline, with a pH of about 7.4 (pH is the degree of acidity or alkalinity on a scale of 1 to 14, with 7 being neutral). In vivo (in the living body), the fluid portion of the blood is called plasma; the cellular portion is referred to as the formed elements. The average adult weighing 70 kg (approximately 154 pounds) has a blood volume of about 5 liters (5.3 quarts), of which approximately 55% is plasma and 45% is formed elements. Accordingly, approximately one half of a blood specimen will be serum or plasma and the other half will be blood cells.

KEY POINT Testing personnel typically prefer specimens that contain roughly 2½ times the amount of sample required to perform the test; so the test can be repeated if needed, with some to spare. Consequently, a test that requires 1 mL of serum or plasma would require a 5-mL blood specimen because only half the specimen will be fluid, while a test that requires 1 mL of whole blood would require a 2½-mL specimen.

Plasma
Normal plasma is a clear, pale-yellow fluid that is nearly 90% water (H2O) and 10% solutes (dissolved substances). The composition of the solute includes the following:
• Gases, such as oxygen (O2), carbon dioxide (CO2), and nitrogen (N).
• Minerals such as sodium (Na), potassium (K), calcium (Ca), and magnesium (Mg). Sodium helps maintain fluid balance, pH, and calcium and potassium balance is necessary for normal heart action. Potassium is essential for normal muscle activity and the conduction of nerve impulses. Calcium is needed for proper bone and tooth formation, nerve conduction, and muscle contraction. In addition, calcium is essential to the clotting process.
• Nutrients, which supply energy. Plasma nutrients include carbohydrates, such as glucose, and lipids (fats), such as triglycerides and cholesterol.
• Proteins, such as albumin, which is manufactured by the liver and functions to help regulate osmotic pressure, or the tendency of blood to attract water; antibodies, which combat infection; and fibrinogen, which is also manufactured by the liver and functions in the clotting process.
• Waste products of metabolism such as blood urea nitrogen (BUN), creatinine, and uric acid.
• Other substances such as vitamins, hormones, and drugs.

Formed Elements

**Erythrocytes**

Erythrocytes (e-ri-th’ro-sites), or red blood cells (RBCs), are the most numerous cells in the blood, averaging 4.5 to 5 million per cubic millimeter of blood. Their main function is to carry oxygen from the lungs to the cells. They also carry carbon dioxide from the cells back to the lungs to be exhaled.

KEY POINT The main component of RBCs is hemoglobin (Hgb or Hb), an iron-containing pigment that enables them to transport oxygen and carbon dioxide and also gives them their red color.

RBCs are produced in the bone marrow. They are formed with a nucleus, which they lose as they mature and enter the bloodstream. Normally a few reticulocytes (re-tik’u-lo-sits), or “retics” (immature RBCs that still contain remnants of material from their nuclear stage), also enter the bloodstream. Mature RBCs have a life span of approximately 120 days, after which they begin to disintegrate and are removed from the bloodstream by the spleen and liver. They are described as anuclear (having no nuclei), biconcave (indented from both sides) disks approximately 7 to 8 microns in diameter. RBCs have intravascular (within blood vessels) function, which means that they do their job within the bloodstream.

**Leukocytes**

Leukocytes, or white blood cells (WBCs), contain nuclei. The average adult has from 5,000 to 10,000 WBCs per cubic millimeter of blood. WBCs are formed in the bone marrow and lymphatic tissue. They are said to have extravascular (outside the blood vessels) function because they are able to leave the bloodstream and do their job in the tissues. WBCs may appear in the
bloodstream for only 6 to 8 hours but reside in the tissues for days, months, or even years. The life span of WBCs varies with the type.

The main function of WBCs is to neutralize or destroy pathogens. Some accomplish this by phagocytosis (fag’o-si-to’sis), a process in which a pathogen or foreign matter is surrounded, engulfed, and destroyed by the WBC. (WBCs also use phagocytosis to remove disintegrated tissue.) Some WBCs produce antibodies that destroy pathogens indirectly or release sub- stances that attack foreign matter.

There are different types of WBCs, each identified by its size, the shape of the nucleus, and whether or not there are granules present in the cytoplasm when the cells in a blood smear are stained with a special blood stain called Wright’s stain. WBCs containing easily visible granules are called granulocytes (gran’-u-lo-sites’). WBCs that lack granules or have extremely fine granules that are not easily seen are called agranulocytes.

Granulocytes
Granulocytes can be differentiated by the color of their granules when stained with Wright’s stain. There are three types of granulocyte: neutrophils, eosinophils (“eos”), and basophils (“basos”). Neutrophils are normally the most numerous type of WBC in adults. A typical neutrophil is polymorphonuclear, meaning its nucleus has several lobes or segments, and is also called a “poly,” “PMN,” or “seg” for short.

Agranulocytes
There are two types of agranulocytes: monocytes (“monos”) and lymphocytes (“lymphs”). Lymphocytes are normally the second most numerous type of WBC and the most numerous agranulocyte. Two main types of lymphocytes are T lymphocytes and B lymphocytes. Monocytes are the largest WBCs.

<table>
<thead>
<tr>
<th>WBC Types</th>
<th>Average Percentage</th>
<th>Description/Staining Characteristics</th>
<th>Function</th>
<th>Life Span</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Granulocytes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neutrophils</td>
<td>65%</td>
<td>Most numerous type of WBC. Segmented or multilobed nucleus. Fine-textured lavender-staining granules.</td>
<td>Destroy pathogens by phagocytosis</td>
<td>6 hours to a few days</td>
</tr>
<tr>
<td>Eosinophils</td>
<td>Up to 3%</td>
<td>Beadlike granules that stain bright orange–red. Two-lobed nucleus.</td>
<td>Ingest and detoxify foreign protein; help turn off immune reactions; increase with allergies and pinworm infestations.</td>
<td>8–12 days</td>
</tr>
<tr>
<td>Basophils</td>
<td>Less than 1%</td>
<td>Least numerous type of WBC: Large dark blue–back staining granules that often obscure a typically S-shaped nucleus.</td>
<td>Release histamine and heparin, which enhance the inflammatory response.</td>
<td>Thought to live several days</td>
</tr>
<tr>
<td><strong>Agranulocytes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Monocytes</td>
<td>1%–7%</td>
<td>Largest WBC: fine, gray–blue cytoplasm and a large, dark staining nucleus.</td>
<td>Destroy pathogen by phagocytosis; first line of defense in the inflammatory process.</td>
<td>Several months</td>
</tr>
<tr>
<td>Lymphocytes</td>
<td>15%–30%</td>
<td>Second most numerous type of WBC: typically has a large, round, dark-purple nucleus that occupies most of the cell and is surrounded by a thin rim of pale-blue cytoplasm.</td>
<td>T lymphocytes directly attack infected cells; B lymphocytes rise to plasma cells that produce immunoglobulins (antibodies) that are released into the bloodstream to circulate and attack foreign cells.</td>
<td>Varies from a few hours to a number of years</td>
</tr>
</tbody>
</table>

**Thrombocytes**

Thrombocytes (throm’bo-sits), better known as platelets, are the smallest of the formed elements. Platelets are actually parts of a large cell called a megakaryocyte (meg’-a- kar’e-o-sit’), which is found in the bone marrow. The number of platelets in the blood (platelet count) of the average adult ranges from 150,000 to 400,000 per cubic millimeter. Platelets are essential to coagulation (the blood-clotting process) and are the first cell on the scene when an injury occurs (see “Hemostasis,” below). The life span of a platelet is about 10 days.

**BLOOD TYPE**
An individual’s blood type (also called blood group) is inherited and is determined by the presence or absence of certain proteins called antigens on the surface of the red blood cells. Some blood-type antigens cause formation of antibodies (also called agglutinins) to the op- posite blood type. Some antibodies to blood-type antigens are preformed in the blood. (A person will not normally have or produce antibodies against his or her own RBC antigens.) If a person receives a blood transfusion of the wrong type, the antibodies may react with the donor RBCs and cause them to agglutinate (a-gloo’tin-ate), or clump together, and lyse (li’s)—that is, to hemolyze or disintegrate. Such an adverse reaction between donor cells and a recipient, which can be fatal, is called a transfusion reaction. The most commonly used method of blood typing recognizes two blood group systems: the ABO system and the Rh factor system.
ABO Blood Group System

The ABO blood group system recognizes four blood types, A, B, AB, and O, based on the presence or absence of two antigens identified as A and B. An individual who is type A has the A antigen, type B has the B antigen, type AB has both antigens, and type O has neither A nor B. Type O is the most common type, and type AB is the least common.

Unlike the ABO system, preformed antibodies in a person’s blood that are directed against the opposite blood type. Type A blood has an antibody (agglutinin) directed against type B, called anti-B. A person with type B has anti-A, type O has both anti-A and anti-B, and type AB has neither. Individuals with type AB blood were once referred to as universal recipients because they have neither A nor B antibody to the RBC antigens and can theoretically receive any ABO type blood. Similarly, type O individuals were once called universal donors because they have neither A nor B antigen on their RBCs, and in an emergency, their blood can theoretically be given to anyone. However, type O blood does contain plasma antibodies to both A and B antigens, and when given to an A or B type recipient, it can cause a mild transfusion reaction. To avoid reactions, patients are usually given type-specific blood, even in emergencies.

Rh Blood-Group System

The Rh blood-group system is based upon the presence or absence of an RBC antigen called the D antigen, also known as Rh factor. An individual with the D antigen present on red blood cells is said to be positive for the Rh factor, or Rh-positive (Rh+). An individual whose RBCs lack the D antigen is said to be Rh-negative (Rh−). A patient must receive blood with the correct Rh type as well as the correct ABO type. Approximately 85% of the population is Rh+.

Unlike the ABO system, antibodies to the Rh factor (anti-Rh antibodies) are not preformed in the blood of Rh− individuals. However, an Rh− individual who receives Rh+ blood can become sensitized. This means that the individual may produce antibodies against the Rh factor. In addition, an Rh− woman who is carrying an Rh+ fetus may become sensitized by the RBCs of the fetus, most commonly by leakage of the fetal cells into the mother’s circulation during childbirth. This may lead to the destruction of the RBCs of a subsequent Rh+ fetus, because Rh antibodies produced by the mother can cross the placenta into the fetal circulation. When this occurs, it is called hemolytic disease of the newborn (HDN).

Compatibility Test/Cross-Match

Other factors in an individual’s blood can cause adverse reactions during a blood transfusion, even with the correct ABO- and Rh-type blood. Consequently, a test to determine if the donor unit of blood and the blood of the patient recipient are compatible (suitable to be mixed together) is performed using patient serum and cells as well as serum and cells from the donor unit. This test is called a compatibility test or cross-match.

BLOOD SPECIMENS

Serum

Blood that has been removed from the body will coagulate or clot within 30 to 60 minutes. The clot consists of the blood cells enmeshed in a fibrin network. The remaining fluid portion is called serum and can be separated from the clot by centrifugation (spinning the clotted blood at a very high speed in a machine called a centrifuge). Normal fasting serum is a clear, pale-yellow fluid. Serum has the same composition as plasma except that it does not contain fibrinogen, because the fibrinogen was used in the formation of the clot. Many laboratory tests, especially chemistry and immunology tests, are performed on serum.

Plasma

Not all tests can be performed on serum. For example, most coagulation tests cannot be performed on serum because some of the coagulation factors (e.g., fibrinogen) are used up in the process of clot formation. Some chemistry tests, such as ammonia and potassium, cannot be performed on serum because clotting releases these substances from the cells. In addition, some chemistry test results are needed stat (immediately) in order to respond to emergency situations; having to wait 30 minutes or more for a specimen to clot before centrifuging it to get serum would be unacceptable. If clotting is prevented, however, coagulation factors and other substances affected by clotting are preserved, and the specimen can be centrifuged immediately. Blood can be prevented from clotting by adding a substance called an anticoagulant. Adding an anticoagulant initially creates a whole-blood specimen. When a whole-blood specimen is centrifuged, it separates into three distinct layers: a bottom layer of red blood cells; a thin, fluffy-looking, whitish-colored middle layer of WBCs and platelets referred to as the buffy coat; and a top layer of liquid called plasma, which can be separated from the cells and used for testing. Normal fasting plasma is a clear to slightly hazy pale-yellow fluid visually indistinguishable from serum. The major difference between plasma and serum is that plasma contains fibrinogen. Many laboratory tests can be performed on either serum or plasma.

Whole Blood

Some tests, including most hematology tests and some chemistry tests such as glycohemoglobin, cannot be performed on serum or plasma. These tests must be performed on whole blood (blood in the same form as it is in the bloodstream). This means that the blood specimen must not be allowed to clot or separate. To obtain a whole-blood specimen, it is necessary to add an anticoagulant. In addition, because the components will separate if the specimen is allowed to stand undisturbed, the specimen must be mixed for a minimum of 2 minutes immediately prior to performing the test.