The Basics of Blood

Fluid

Blood has two primary components—fluid and cells. When the blood is in the body, its fluid is called "plasma." Plasma suspends blood cells and helps transport them and other nutrients throughout the body. It also helps remove waste. Plasma is about 90% water, and the rest consists of products like protein molecules and electrolytes.

The electrolytes in plasma aid the body with chemical reactions and maintaining stable blood pH levels. The proteins in plasma do different jobs, primarily fighting infection and assisting with coagulation, or blood clotting. When blood clots, the proteins that assist the clotting (clot proteins) are removed from the plasma, and the clot is formed.

After the clotting process, the fluid part of blood is called "serum" because it no longer contains the clot proteins. Similarly, when a blood sample is put in a tube that does not contain an anticoagulant, the blood follows it natural clot process, and, after centrifugation, becomes serum. If blood is collected in a tube with an anticoagulant, it will not clot, and the fluid will still be plasma.

The most common anticoagulant used for hematology—the study of how blood functions—testing is EDTA, which is identified in practice by a purple- or lavender-topped tube. Other anticoagulants are used for other types of testing, like internal organ chemistries.

When blood is in the body or mixed with an anticoagulant, it is called "whole blood;" when blood has clotted, it is referred to as clotted blood or serum. When a sample is placed in a tube without anticoagulant, it is because serum is needed for testing. In some cases, plasma is used for testing.

Photos courtesy of Alexandria Stone

Figure 1: Clotted serum separator tube (SST) before spinning.

Figure 2: Loaded swivel head centrifuge with SST tubes.

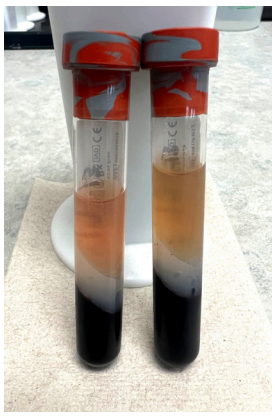


Figure 3: Spun SST: mild hemolysis on the left and no hemolysis on the right.

Cells

There are three main types of blood cells: erythrocytes (or red blood cells), leukocytes (or white blood cells) and thrombocytes (or platelets).

Red Blood Cells

Erythrocytes, or red blood cells (RBCs), are the most numerous of the blood cells. They have a biconcave (curving inward on both sides) disc shape—like a doughnut without a hole—in most mammals. (In birds and reptiles, the cell shape is more oval.) The concave areas in the middle of the cell are where hemoglobin, the protein inside RBCs that give them their red color, carries oxygen. The hemoglobin within RBCs has an extremely vital task: to transport fresh oxygen to tissues throughout the body and to remove carbon dioxide from the body by carrying it to the lungs to be exhaled. If the hemoglobin in RBCs is damaged (for example, by exposure to excessive carbon monoxide), it cannot perform its job, and the body will suffer a low oxygen level (hypoxia). Without proper, healthy, oxygenated blood flow, animal cells die.

RBCs are made in the bone marrow and then released into the bloodstream, and their shape reflects their maturation process. When they are young (before they are released into the bloodstream), they still contain nuclei. The last step in the maturation process is for the cell to push out the nucleus, which gives them their biconcave shape. Sometimes, however, red blood cells need to be released early (think serious injury or a disease like hemolytic anemia that destroys red blood cells faster than they can be made). In such cases, RBCs will still contain some of their nuclear remnants and have a different appearance on a blood smear.

White Blood Cells

Leukocytes, or white blood cells (WBCs), are the next most numerous blood cell. There are five different kinds of white blood cells, and each one performs different jobs in the body. These five types of WBCs are also further categorized as either granulocytes or agranulocytes.

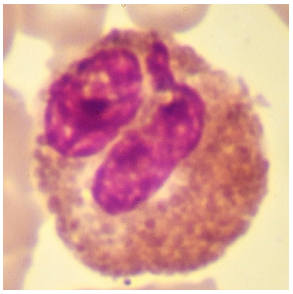
Granulocytes contain granules in their cytoplasm—cytoplasm is the jelly-like substance inside cells, but outside the nucleus; it's made of water, salts and different organic molecules—that perform different jobs according to the type of WBC they're part of. Agranulocytes do not contain granules in their cytoplasm.

The following three types of white blood cells are granulocytes:

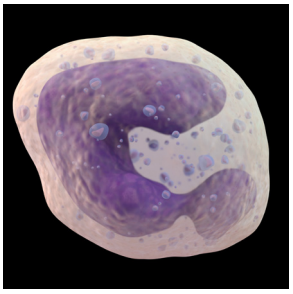
- **Neutrophils** are the first white blood cells released in response to injury or infection. Their purpose is to stimulate inflammation and phagocytize (ingest; surround and destroy; eat) bacteria and other harmful particles while also signaling other blood cells to start their jobs (if needed). Their activity and death in large numbers forms pus.[103]


jarun011/Adobe Stock
Neutrophil

- **Eosinophils** are usually seen in higher amounts in the circulating blood when the body is experiencing an allergic reaction like a reaction to a bee sting or allergic reaction to food. Eosinophils are also seen with parasitic infections. Eosinophils are easily identified because granules in their cytoplasm appear as having pink dots when stained.[104]

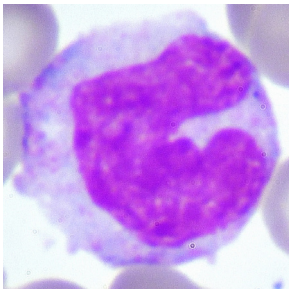

jarun011/Adobe Stock
Eosinophil

- **Basophils** take part in allergic reactions, especially anaphylaxis, as they release the chemical histamine. Once stained, basophils are characterized by their large blue granules.

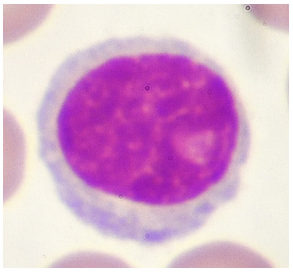

gaetan/Adobe Stock
Basophil

The following two types of white blood cells are agranulocytes:

- **Lymphocytes**—There are two types of lymphocytes: B-cells and T-cells. B-cells make infection-fighting proteins called antibodies in response to specific antigens (a toxin or foreign substance that signals the immune system to respond), a process called cellular immunity. B-cells are also capable of remembering the pathogens they respond to and passing that information to future generations of B-cells, a process called humoral immunity. T-cells help signal and activate B-cells. Lymphocytes are identified by a large dense nucleus that stains dark purple.


jarun011/Adobe Stock
Lymphocyte

- **Monocytes**—Monocytes circulate in the veins and arteries until later in their lives when they move into body tissues and are then called macrophages. (Monocytes and macrophages are not two different types of white blood cells; macrophage is the new name used for monocytes in a later stage of life.) Like neutrophils, macrophages phagocytize bacteria and other harmful particles, but they do this without creating inflammation.


jarun011/Adobe Stock
Monocyte

Platelets

Thrombocytes, or platelets, are not full cells. They are fragments of larger cells called megakaryocytes. Megakaryocytes release fragments with "sticky arms" into circulation. These fragments are platelets (or thrombocytes), and they are activated by the coagulation process described earlier in the section titled *Fluid.* Once the platelets get the signal, they rush to the site of bleeding, attach themselves to each other, and combine with coagulation proteins called "fibrin" to form a "platelet plug," which ultimately stops the bleeding if the injury is small enough for the body to handle on its own.

The Complete Blood Count (CBC)

The complete blood count, commonly referred to as the CBC, is a series of hematological tests that determine the total number and types of blood cells present in a given amount of a patient's blood. This is one of the more common laboratory tests performed to reveal abnormalities of the blood. The tests that make up the CBC may be completed at an off-site laboratory or on-site at the veterinary facility. Certain tests, however, are typically completed on-site before samples are sent to the laboratory. While the veterinary technician completes these tests, ideally, the veterinary assistant is also familiar with them as well.

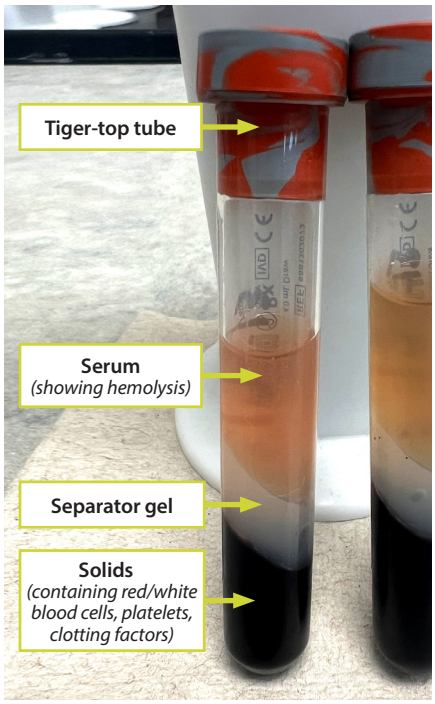


Photo courtesy of Alexandria Stone

Separator gel helps keep the serum and packed RBCs separate after centrifugation.

The tests that make up the CBC include:

- Total red blood cell (RBC) count
- PCV (packed cell volume) (% of total blood volume)
- Total white blood cell (WBC) count
- RBC indexes including
  - Mean corpuscular volume (MCV)—average volume or size of red blood cells[105]
  - Mean corpuscular hemoglobin (MCH)—mean weight of hemoglobin (Hgb) the average red blood cell contains
  - Mean corpuscular hemoglobin concentration (MCHC)—average concentration of hemoglobin in the average red blood cell

- Hemoglobin (Hgb) concentration—Hgb is an iron-protein that binds and carries oxygen.
- Actual platelet determination
- Reticulocyte count (when the patient is anemic)—Reticulocytes are immature RBCs that contain RNA (Ribonucleic acid that is involved with synthesizing protein and may contain DNA) that is not present in mature RBCs. Reticulocyte numbers increase when bone marrow responds to an anemia.
  - **NOTE:** *Reticulocytes are RBCs that were released early (because of injury or infection) and still retain remnants of their nuclei. This type of RBC is has a different appearance on a blood smear, and if a typical three-part Wright's stain is used, they are called "polychromatophils," but if methylene blue is used, they are called "reticulocytes."*
  - Anemia—a deficiency in the number, size and oxygen-carrying abilities of RBCs.[106]

To view a short video showing the steps of a CBC, please log into the Student Center, click Training Videos and choose *Complete Blood Count.*

The examination of blood is performed from a stained blood smear in the monolayer (or single layer) or the feathered edge. The slide is viewed on the oil-immersion (100x) objective of the microscope. When studying the form and structure of red blood cells, there are several items that must be observed and recorded to help in diagnosing what's normal and what is abnormal:

- RBC morphology—information related to the form and structure (size, shape, color, etc.) of the cells
- Differential (different) WBC count—counts of neutrophils, eosinophils, basophils, lymphocytes and macrophages. Eosinophils appear as having pink dots when stained; basophils are identified by their large blue granules; lymphocytes have a large dense nucleus that stains dark purple.
- Platelet estimation—Platelets are tiny, and their cytoplasm stains purple. Increased platelet numbers may be seen with blood loss. More commonly, a lack of platelets indicates immune problems that mainly effect blood clotting.

Laboratory Testing Safety Precautions

- Always wear examination gloves if the patient is suspected of having a zoonotic disease.
- Make sure to label all samples with the patient's identifying information such as name, owner's name, patient ID number (if applicable), and species.
- Become familiar with all laboratory equipment before attempting to perform tests.
- Follow all manufacturers' instructions precisely.
- Be cautious when using laboratory equipment; it is expensive and can be hard to replace.
- Close sample containers immediately after inserting or removing laboratory samples.
- Do not keep open sample containers near each other, as this can cause cross-contamination.
- Make sure all laboratory request forms are filled out completely to avoid patient misidentification (if samples are being sent out to a commercial lab).
- Refrigerate samples if they are not being immediately tested.
- Ensure that all samples are picked up by the off-site laboratory before the end of the day to avoid their expiration.
- Check expiration dates on reagents.
- Record any laboratory results in the patient's record immediately.
- Make sure any blood-contaminated materials are disposed of in a biohazard container.