Specimen Collection

Laboratory tests contribute vital information about a patient's health. Correct diagnostic and therapeutic decisions rely, in part, on the accuracy of test results. Adequate patient preparation, specimen collection, and specimen handling are essential prerequisites for accurate test results. The accuracy of test results is dependent on the integrity of specimens.

Safety and Disposal Considerations in Specimen Collection

In all settings in which specimens are collected and prepared for testing, laboratory and health care personnel should follow current recommended sterile techniques, including precautions regarding the use of needles and other sterile equipment. Treat all biological material as material that is potentially hazardous as well as contaminated specimen collection supplies. For all those who are involved in specimen collection and preparation, the responsibility to adhere to current recommendations designed to maintain the safety of both patients and health care workers does not end when the patient is dismissed.

There are four steps involved in obtaining a good quality specimen for testing:

(1) preparation of the patient.

(2) collection of the specimen.

(3) processing the specimen.

(4) storing and/or transporting the specimen.

Preparation

Prior to each collection, review the appropriate test description, including the specimen type indicated, the volume, the procedure, the collection materials, patient preparation, and storage and handling instructions.

Preparing the Patient. Provide the patient, in advance, with appropriate collection instructions and information on fasting, diet, and medication restrictions when indicated for the specific test.

Preparing the Specimen. Verify the patient's identification. Proper identification of specimens is extremely important. All primary specimen containers must be labeled with at least two identifiers at the time of collection. Submitted slides may be labeled with a single identifier, but two identifiers are preferred. Examples patient's name (patient's first and last name exactly as they appear on the test request form), date of birth, hospital number, test request form number, accession number, or unique random number. A location such as a hospital room number is not an appropriate patient identifier. All specimens should be labeled in the presence of the patient

Blood Collection

There are two types of blood samples: venous blood and capillary blood.

Venous Blood: Veins are the most commonly used site for blood collection. The process of drawing blood from a vein is known as venipuncture. As a general rule, arm veins are the best source of blood. The preferred site is the antecubital fossa, which is where the arm bends at the elbow. The vein of choice is the median cubital vein. The cephalic and basilic can also be used. In unusual situations, it may be necessary to use hand or wrist veins when a suitable arm vein cannot be located (e.g. obese patients), or if the arms are bandaged or have been punctured repeatedly

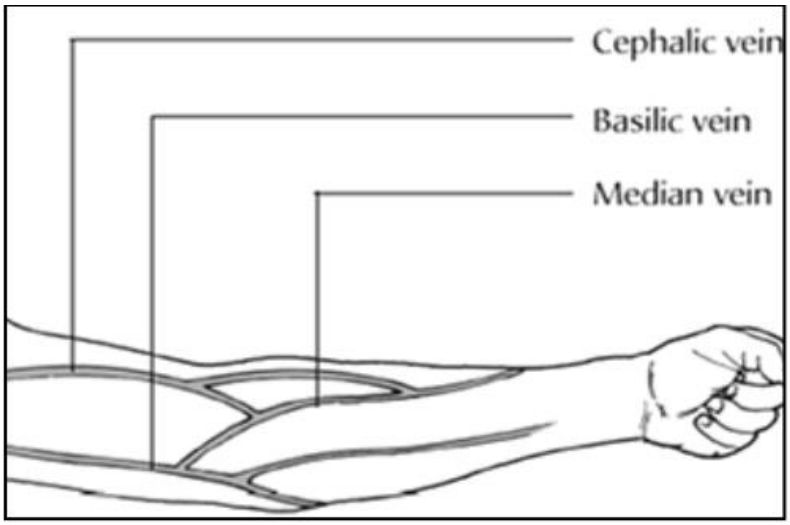


Figure 1: Common arm veins used for venipuncture.

Venipuncture Procedure

1. Ask the patient to make a loose fist and Select the appropriate vein for venipuncture.

2. Apply the tourniquet 3-4 inches above the collection site.

3. Clean the puncture site by making a smooth circular pass over the site with a 70% alcohol pad, moving in an outward spiral from the zone of penetration.

4. Uncap the needle with the bevel pointing upward.

5. Pull the skin tight with your thumb or index finger just below the puncture site.

6. Holding the needle in line with the vein, use a quick, small thrust to penetrate the skin and enter the vein in one smooth motion.

7. Holding the needle hub securely, insert the first vacutainer tube into the tube connector. Blood should flow into the vacuum tube.

8. After blood starts to flow, release the tourniquet and ask the patient to open his or her hand.

9. Once tube is full, use one hand to stabilize the needle in the vein and remove the tube with the other.

10. Insert other tubes in the correct order until all test samples are obtained.

11. Place a gauze pad over the puncture site and remove the needle. Immediately apply slight pressure, and then apply a fresh bandage over the penetrating site.

Needles Used for Venipuncture

- 19 or 21 gauge for adults.
- 23 gauge for children.

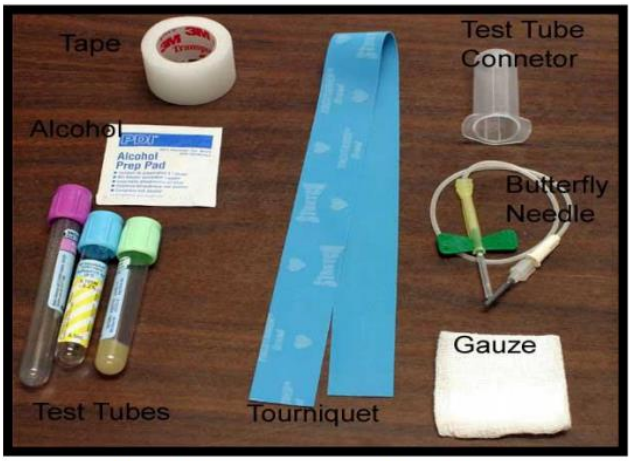
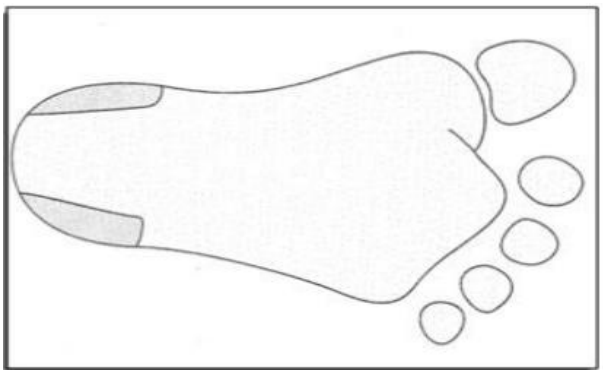


Figure 2: Common tools used for phlebotomy

Capillary Blood

Capillary blood is used for infants under 1 year of age and when it is not possible to obtain venous blood. Common sites for drawing capillary blood are the heel and the ball of the middle finger. Skin puncture is carried out with a needle or lancet. In adults and older children blood can be obtained from a finger. The recommended site is the distal digit of the third or fourth finger on its palmar surface, about 3-5 mm lateral from the nail bed. Formerly, the earlobe was commonly used, but it is no longer recommended because reduced blood flow renders it unrepresentative of the circulating blood. In infants, satisfactory samples can be obtained by a deep puncture of the plantar surface of the heel .Because the heel should be very warm, it may be necessary to bathe it in hot water. Capillary blood is free flowing, and thus it causes a greater risk of contamination and disease transmission. Venous blood is therefore more convenient for handling and it yields considerably more accurate results.



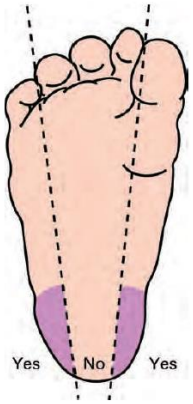


Fig. 1.5 The areas of the foot of a baby or infant that are suitable for obtaining capillary blood.

Collection of Capillary Blood

1. Clean the area with 70% alcohol and allow to dry.

2. Puncture the skin to a depth of 2-3 mm with a sterile lancet.

3. Wipe away the first drop of blood with dry sterile gauze.

4. Collect the second and following drops onto a reagent strip or by a 10 ml or 20 ml micropipette.

Why Is a Blood Test a Good Measure of Health?

Bloodwork, shows the actual physiological response and status in the body to different interventions. Routine blood testing is one of the most reliable and important tools to help you evaluate and track your physical health over time.

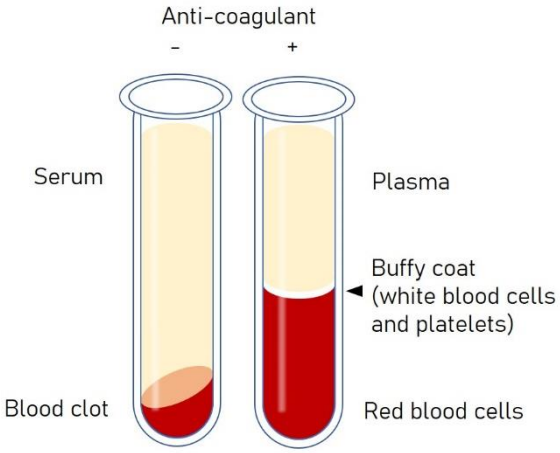
Blood Parts

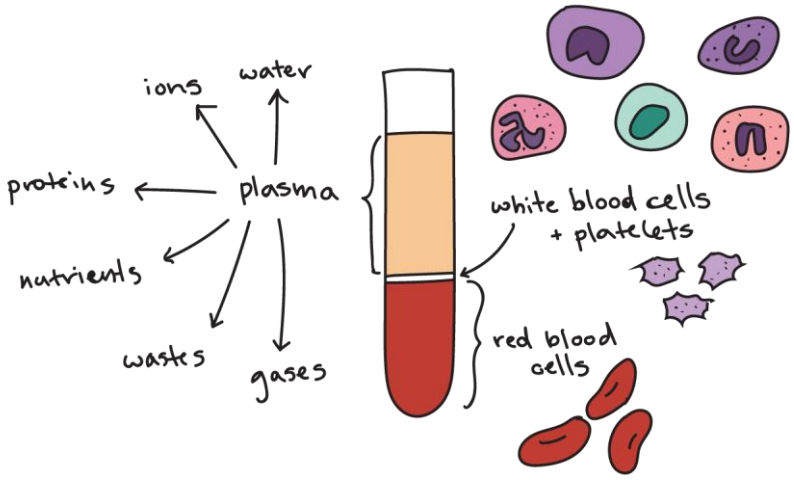
Blood is composed of two parts: a cellular part (blood cells) and a non cellular part called plasma. Blood Cells There are three types of blood cells: Red blood cells, white blood cells (leukocytes) and platelets.

Plasma; Plasma is the clear yellowish portion of blood. It contains fibrinogen, a coagulation factor that causes plasma to clot quickly. To obtain plasma, blood is collected into a tube with an anticoagulant.

Serum; Serum is the part of the blood that contains neither blood cells nor clotting factors. Serum is prepared by allowing the blood to naturally form a clot, and then using a centrifuge to remove the blood cells and the clot. The upper supernatant after centrifugation is the serum. To obtain serum blood should be collected into a sterile tube that contains no anticoagulants.

Figure4: Vacutainer tube for serum. It contains no anticoagulants





Anticoagulants: Whole blood is necessary for most haematological tests. Blood samples must be collected into tubes or bottles containing different anticoagulants to prevent coagulation (clotting).

The anticoagulants commonly used are

Ethylenediaminetetraacetic Acid (EDTA), Sodium Citrate and Heparin.

Ethylenediaminetetraacetic Acid is a chelating agent produced as a series of salts. The sodium and potassium salts EDTA are powerful anticoagulants, and they are especially suitable for routine haematological work.

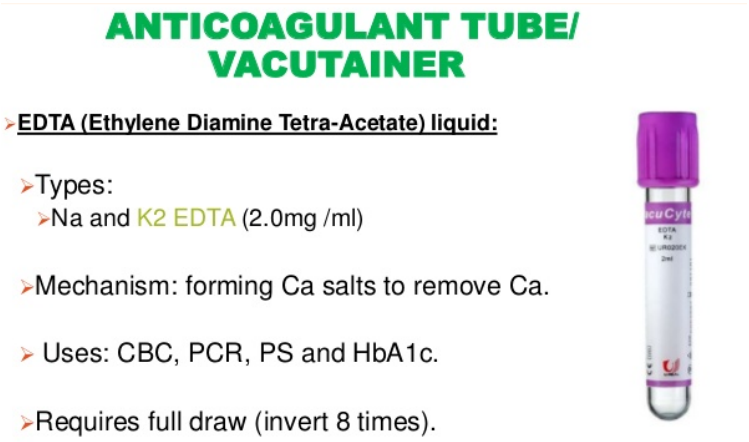


Figure 5: EDTA blood collection tube.

Chemical Action EDTA acts by its chelating effect on the calcium molecules in the blood. It removes calcium ions, which are essential for coagulation.

Use EDTA is the anticoagulant of choice for blood counts and blood films. It is also ideal for platelet counts as it prevents platelets from clumping. This anticoagulant, however, is not suitable for coagulation studies because it destroys clotting factors V and VIII.

Concentration The recommended concentration of EDTA is 1.5 ± 0.25 mg/ml of blood. Excess of EDTA affects both red cells and leukocytes causing shrinkage and degenerative changes. A concentration exceeding 2 mg/ml of blood may result in a significant decrease in packed cell volume (PCV) and an increase in mean cell hemoglobin concentration (MCHC). The platelets are also affected; excess of EDTA causes them to swell and then disintegrate, leading to an artificially high platelet count. Therefore, it is important to ensure that the correct amount of blood is added and that the anticoagulant is thoroughly mixed in the blood added to it.

Trisodium Citrate Chemical Action: Trisodium citrate removes free calcium ions by loosely binding to them forming a calcium citrate complex. This anticoagulant is used for coagulation studies and the estimation of the erythrocyte sedimentation rate (ESR).

Concentration For coagulation tests, nine volumes of blood are added to one volume of the sodium citrate solution. For the ESR, four volumes of blood are added to one volume of the sodium citrate solution. Sodium citrate is available in two concentrations: 3.2% and 3.8%.

Figure 6: Sodium citrate blood collection tube

- **BLACK:**
- Na citrate 1:4.
- 3.8% of sodium citrate
- Action: Remove calcium.
- Uses: Westergren – Erythrocyte Sedimentation Rate (ESR).

Sodium citrate :

- (1:9 ratio).
- Anticoagulant: 3.2%
- Mechanism: Calcium chelation.
- Use: Coagulation studies and platelet function.

Heparin: Heparin is a natural substance which is synthesized by the liver. Chemical Action Heparin neutralizes thrombin, an essential clotting factor, with the aid of a co-factor present in the albumin fraction of plasma.

Use Heparin is an effective anticoagulant and it does not alter the size of red cells. It serves as a good dry anticoagulant when it's important to minimize the chance of lysis after blood has been withdrawn.

Heparin is the best anticoagulant for osmotic fragility and is suitable for immunophenotyping. Heparinized blood is not recommended for cell counting because of its clumping effect on platelets and leukocytes. It also should not be used for making blood films because it gives a faint blue coloration to the background when Romanowsky dyes are used. It inhibits enzyme activity, and it is not suitable for use in the study of polymerase chain reaction (PCR) with restriction enzymes.

Concentration the heparin concentration used

is 10-20 international units (IU) per Ml of blood.

Figure7: Heparin Blood Collection

Red Plain Tube; No preservative or anticoagulant. Uses; usually for toxicology and serology

SST (No additives), Clotting accelerator and separation gel, uses for chemistry, Immunology and Serology.

PST/ Light Green:- Plasma Separating Tube with Lithium Heparin

Advantage: Forms aphysical barrier between plasma and blood cells during centrifugation.