I. **Blood Collection and Anticoagulants**

A. Laboratory tests can be done on the cellular or fluid portions of the blood. The use of different blood collection tubes *with or without an anticoagulant* determines the portion of the blood which can be analyzed.... **whole blood, plasma, or serum.**

1. In *vivo*, normal blood should not clot unless an injury occurs. Upon injury, coagulation reactions involving platelets and plasma clotting proteins transform fluid blood into a clot at the site of injury.

2. In *vitro*, freshly drawn blood is fluid, but within minutes will spontaneously clot. Clotting is triggered by surface contact (tube) or poor blood drawing technique.

B. **Coagulation reaction:**

   **Platelets & Clotting proteins + calcium \(\rightarrow\) thrombin which converts fibrinogen \(\rightarrow\) fibrin**

1. Fibrin strands trap RBCs, WBCs, and PLTs in the clot.
2. Fibrinogen and other clotting proteins are "used up" in the formation of fibrin.
   - **Cells and clotting proteins cannot be measured if coagulation occurs.**
3. To prevent activation of blood coagulation, **anticoagulants** are used.

C. **Anticoagulant** - a chemical substance that *irreversibly* prevents blood coagulation by:

1. Removing/binding calcium – EDTA and sodium citrate
2. Inactivating thrombin - heparin
3. Blood collection tubes are color-coded and labeled with the type of additive (powder or liquid) in the tube. Some tubes used:
   a. Red top "clot" tube - contains no anticoagulant/additive…the blood will clot.
   b. Purple top tube - contains EDTA anticoagulant to prevent clotting.
   c. Blue top tube - contains sodium citrate anticoagulant to prevent clotting.
   d. Green top tube – contains heparin anticoagulant to prevent clotting.

D. To obtain whole blood or plasma - blood is drawn in a tube containing an **anticoagulant**:

1. Clotting is prevented if the blood is properly drawn and mixed with the anticoagulant.
2. After mixing, the cells of **whole blood** can be analyzed.
3. Centrifugation of whole blood separates the cells from the fluid plasma.
4. After centrifugation, the fluid **plasma** can be analyzed. Plasma contains fibrinogen.

E. Spontaneous clotting occurs if blood is drawn in a tube with **no anticoagulant**:

1. Fibrinogen is used to form fibrin strands which trap cells in the blood clot.
2. Centrifugation separates the clot from the fluid serum.
3. After centrifugation, the fluid **serum** can be analyzed. Serum lacks fibrinogen.
F. To analyze blood constituents, each laboratory area has specific specimen requirements:
   1. Hematology
      a. Cell counts (such as platelet counts) are performed on whole blood drawn in EDTA (purple top) which is best for preserving the cellular elements.
      b. Coagulation studies (PT or APTT) are performed on plasma drawn in liquid sodium citrate (blue top) which is best for preserving the coagulation proteins.
   2. Chemistry - tests are performed on plasma drawn in heparin or on serum.

II. Sources of Blood Collection Error

The 1st step in obtaining accurate test results is the proper collection of the blood specimen. Specimens unsuitable for testing must be recognized and redrawn to avoid pre-analytical errors.

A. Wrong patient identification/labeling error!

B. Use of wrong anticoagulant/tube – unable to perform the ordered test; blood must be redrawn.
   1. Heparin is unacceptable for coagulation tests…causes falsely prolonged results.
   2. Heparin causes platelet clumping so platelet counts cannot be done.

C. Hemodilution – if blood is drawn above an IV line, the specimen is diluted out causing false low cell counts and/or is contaminated.

D. Hemoconcentration - prolonged tourniquet time (>1 min) restricts blood flow causing false high cell counts.

E. Wrong ratio of blood to anticoagulant - all tubes have minimum draw amounts to maintain the proper anticoagulant concentration to blood volume, e.g. blue top tubes for coagulation tests must be full.

F. Hemolysis - rupture of blood cells causes release of cell constituents such as potassium, LD, and tissue factor. Hemolysis is caused by traumatic blood drawing technique, forcing blood through the syringe needle into tube, or vigorous shaking of the blood tube rather than tilting. Always TILT the tube when mixing the blood with the anticoagulant additive.

G. Partially clotted blood draws - blood which is drawn too slowly or not mixed properly with anticoagulant-containing tubes may clot.
   1. Cells are trapped in the fibrin clot causing falsely low cell counts.
   2. Certain coagulation proteins are removed causing falsely prolonged test results.

H. Specimen handling requirements must be followed, e.g., certain tests have time limits for testing.