Department of Biochemistry

Test Tube & Pipette

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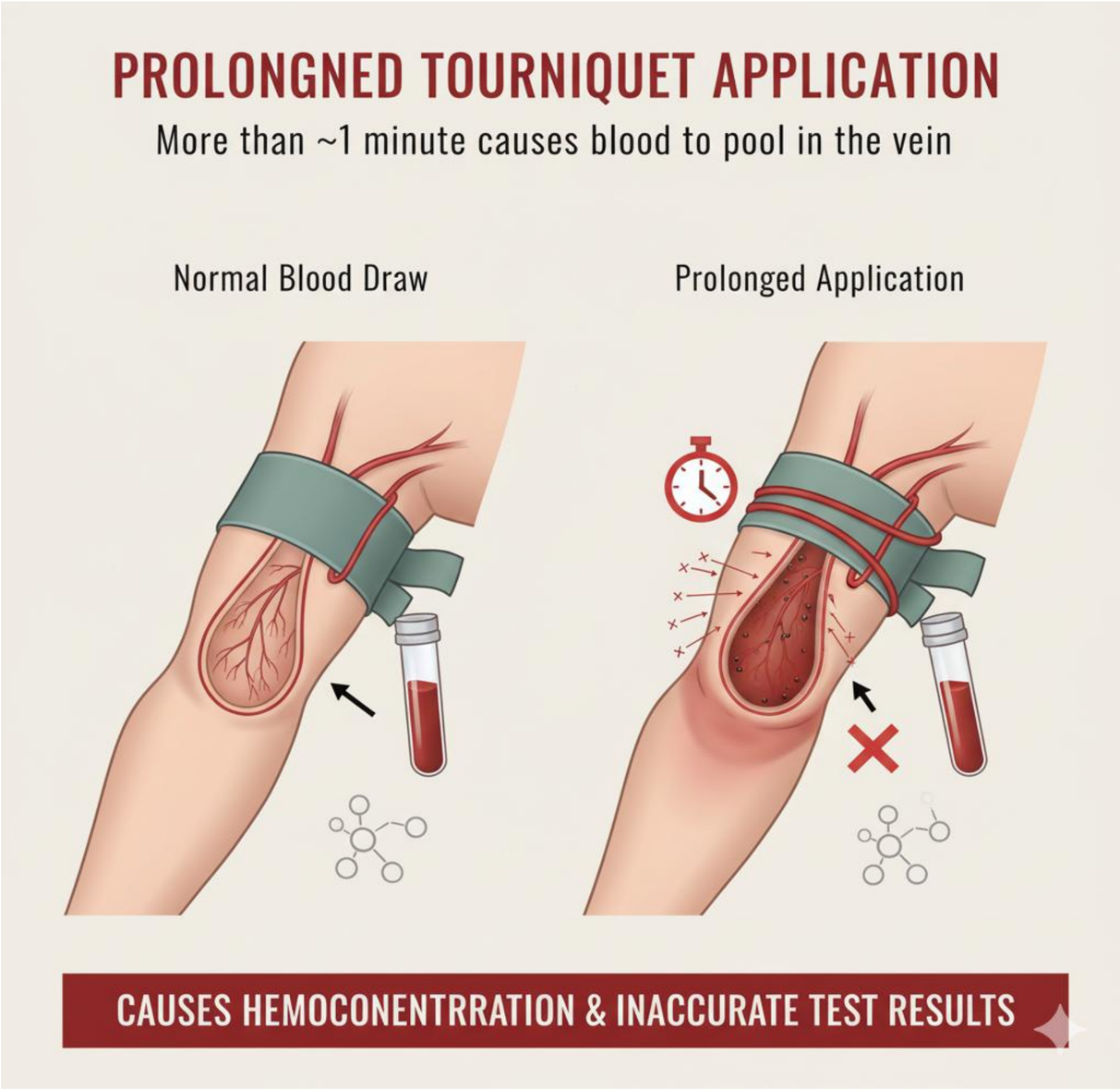
LEARNING OBJECTIVES

- Understand the Types of Test Tubes
- What is a micropipette
- Application of micropipette
- How to use a micropipette

It is not recommended to use the tourniquet for more than one minute in certain tests, especially for potassium and calcium. **For potassium**, pressure may cause partial hemolysis or potassium leakage from red blood cells, and **for calcium**, hemoconcentration raises plasma proteins (especially albumin), which increases the measured total calcium .At the same time, changes in pH during venous stasis can also affect ionized calcium levels.



Green-top tube (lithium heparin):

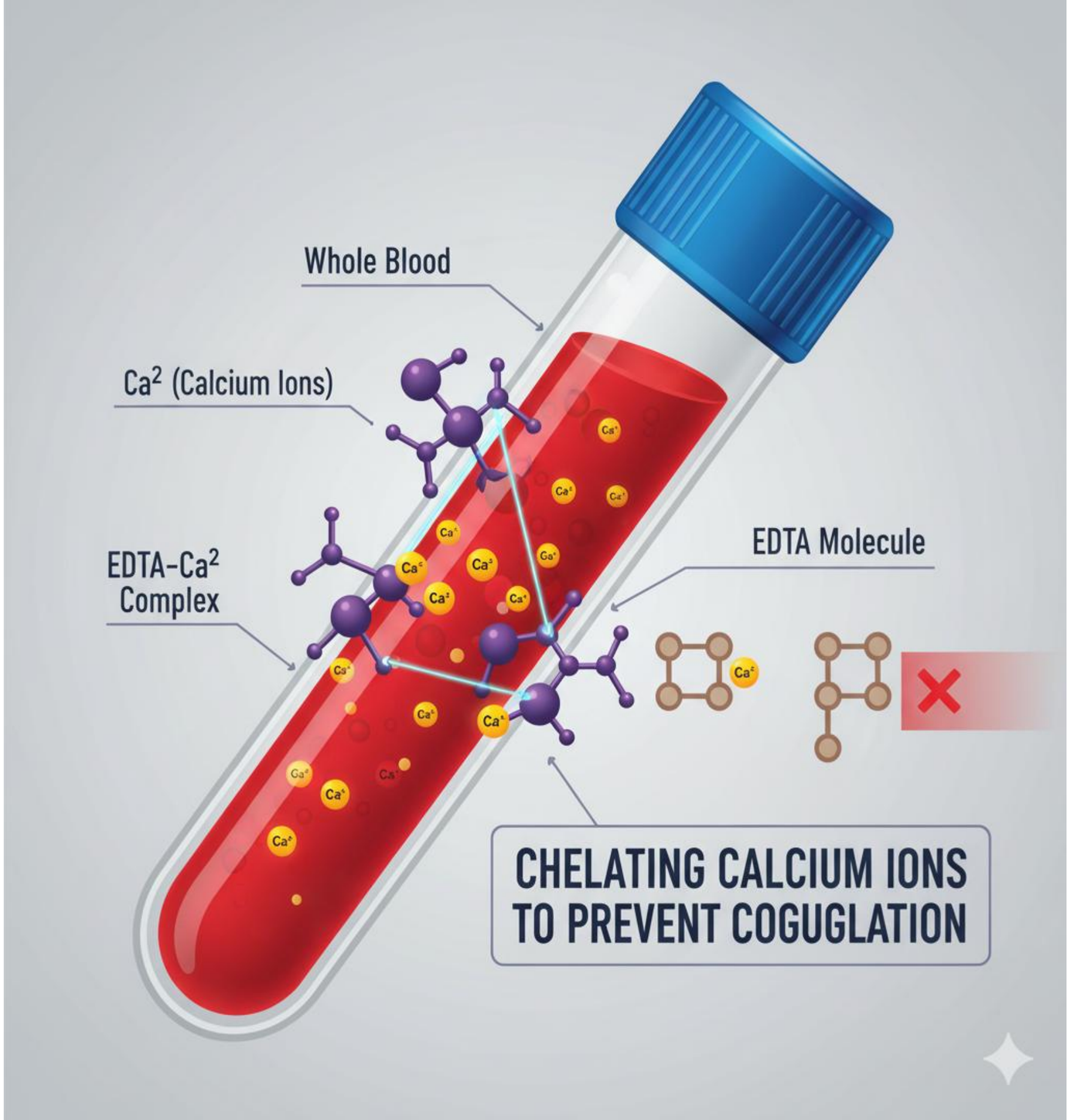
1. Tube contains lithium heparin as an anticoagulant.
2. This tube is used for preparing heparinized plasma, whole blood, and bone marrow specimens.
3. Used for arterial blood gas analysis, plasma electrolyte determination
4. Heparin prevents coagulation by activating antithrombin III, which inhibits thrombin (factor IIa) and factor Xa

Pink, Lavender-top tube (EDTA):

1. Tube contains K3-EDTA as an anticoagulant.
2. EDTA: Ethyl Diamine Tetra Acetic Acid
3. This tube is used for preparing whole blood and bone marrow specimens.
4. Used in complete blood count (CBC), HbA1c

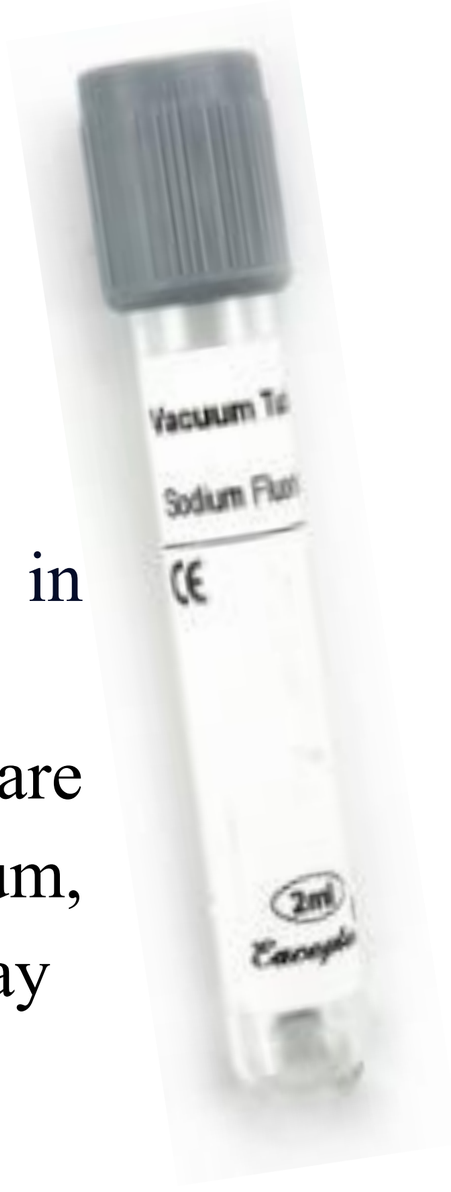
Advantage

preservation of cellular components and the morphology of blood cells

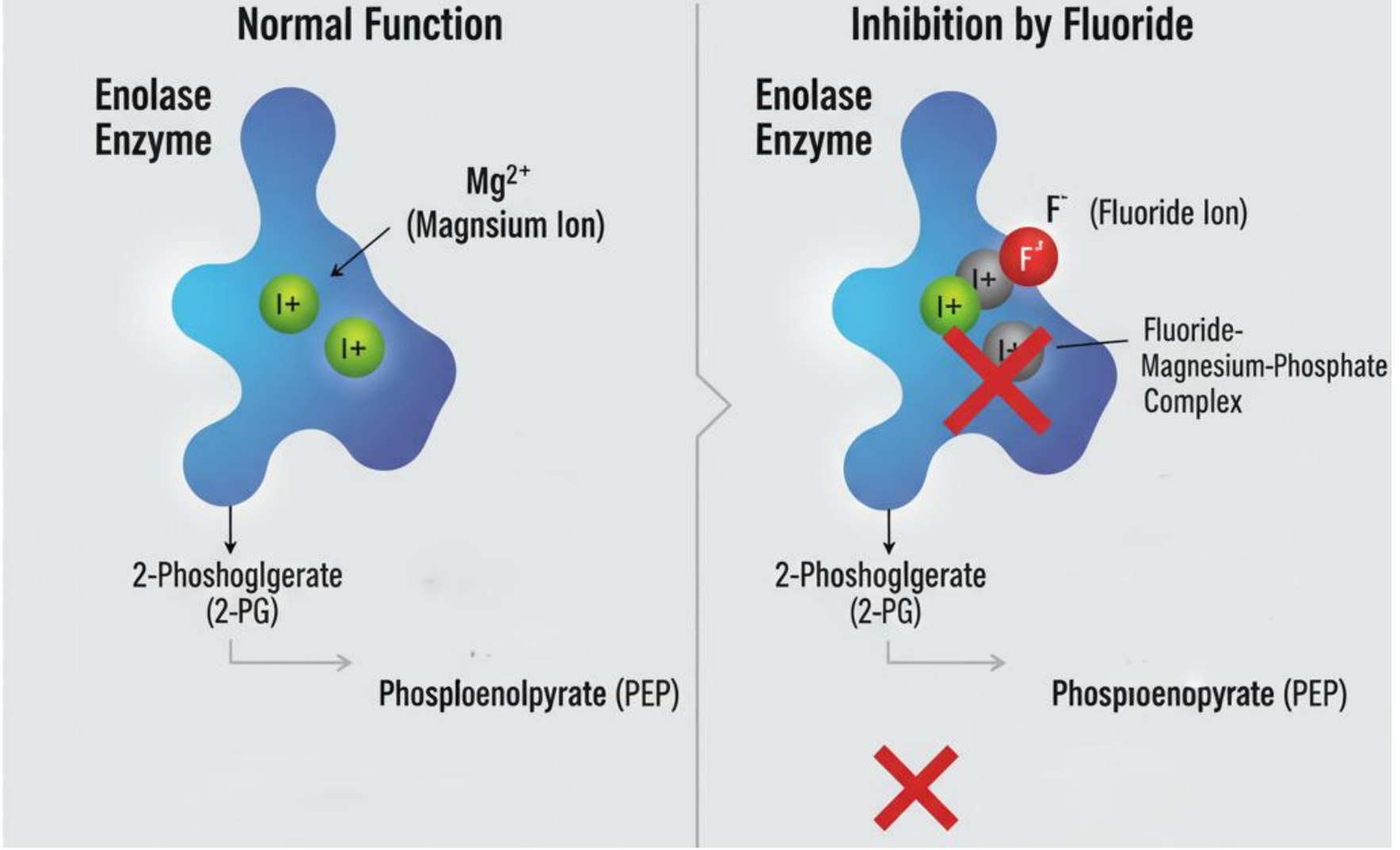


Gray tube (oxalate fluoride)

1. Tube contains a weak anticoagulant
2. It contains potassium oxalate/sodium fluoride
3. This tube is used for collecting plasma.
4. It is used for Fasting Blood Sugar, the lactate test
5. Fluoride works as an anti-glycolytic that inhibits glycolysis in erythrocytes, stabilizing glucose levels in the tube
6. Fluoride forms a stable complex with magnesium ions, which are essential cofactors for the enzyme **Enolase**. By binding to magnesium, fluoride inhibits Enolase activity, thereby blocking glycolytic pathway



FLUORIDE INHIBITION OF ENOLASE

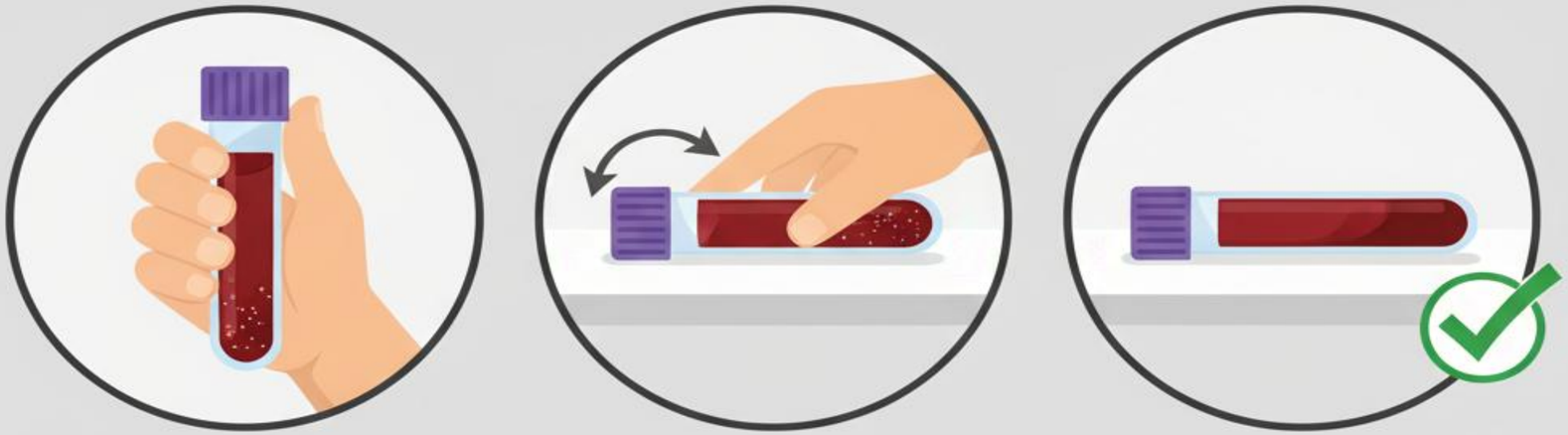


Light blue-top tube (sodium citrate):

1. Tube contains sodium citrate as an anticoagulant.
2. This tube is used for preparing citrated plasma for coagulation studies.
3. Used for PT, PTT, and Fibrinogen test
4. Sodium citrate binds calcium ions (Ca^{2+}) in the blood, since calcium is essential for activating the coagulation cascade..
5. By binding calcium, it temporarily prevents clotting.

PROPER TUBE ROLLING FOR ANTICOGULATION

Ensuring homorgeus mixing by gentle side-side rolling to prevent cloting and maintain sample integrity



1. Fill Tube

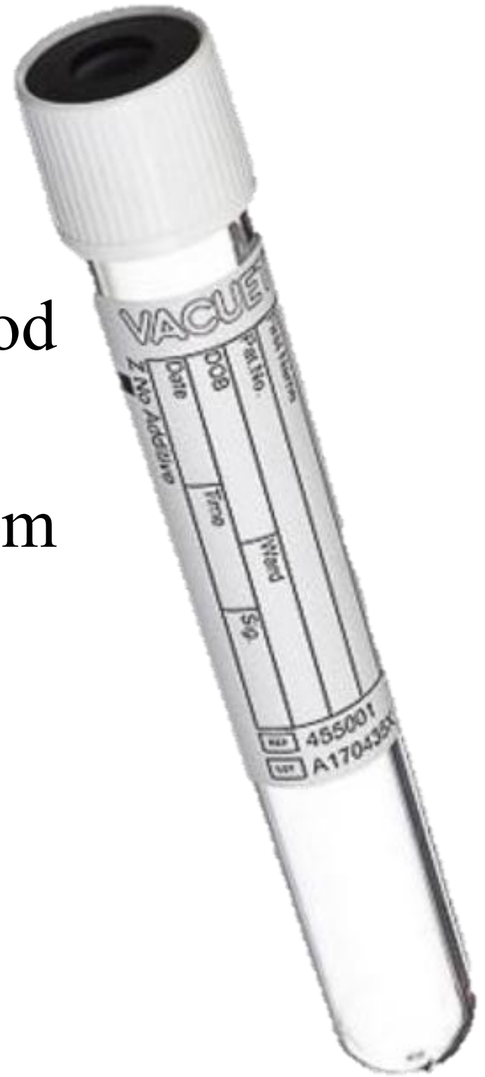
2. Gentle Side-Stide Rolling (5-10x)

3. Fully Mixed (No Clot)

PREVENT CLOTTING & ENSURE ACCURATE TEST RESULTS

White or red-top tube: (Plain tube)

1. Tube **does not** contain an anticoagulant.
2. This tube is used for collecting serum or clotted whole blood specimens.
3. The blood is allowed to clot for at least 10–15 minutes at room temperature and then centrifuged for 15 minutes to get serum
4. Used only to obtain serum

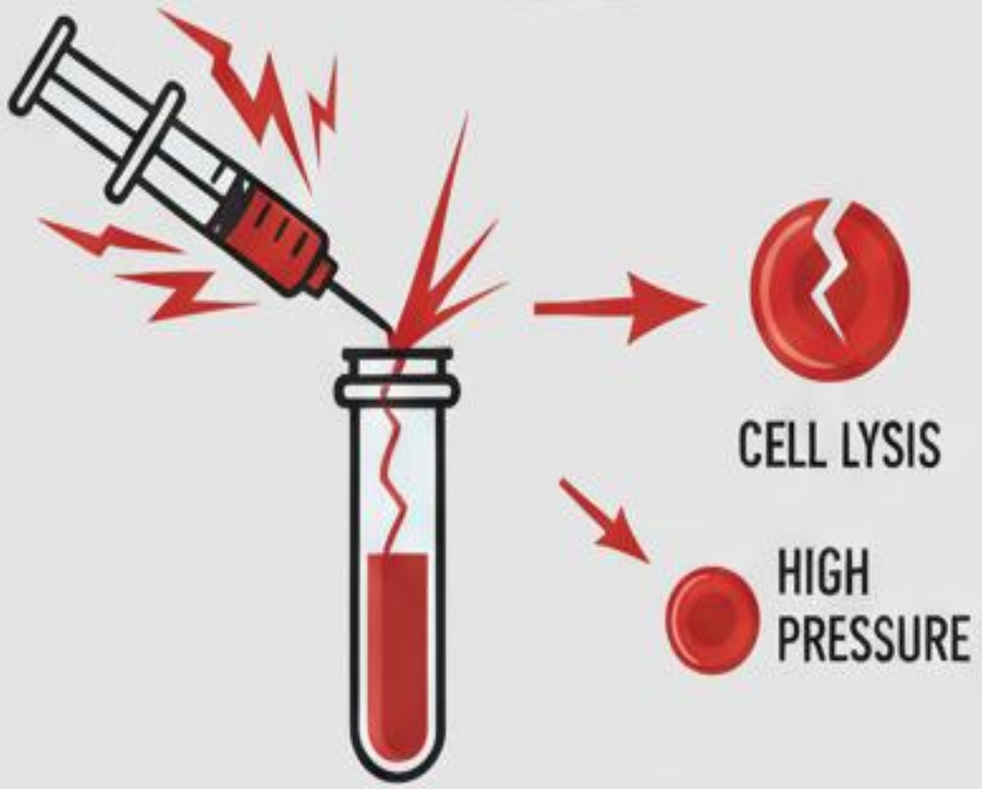


Tiger-top tube (serum separator)(gel tube):

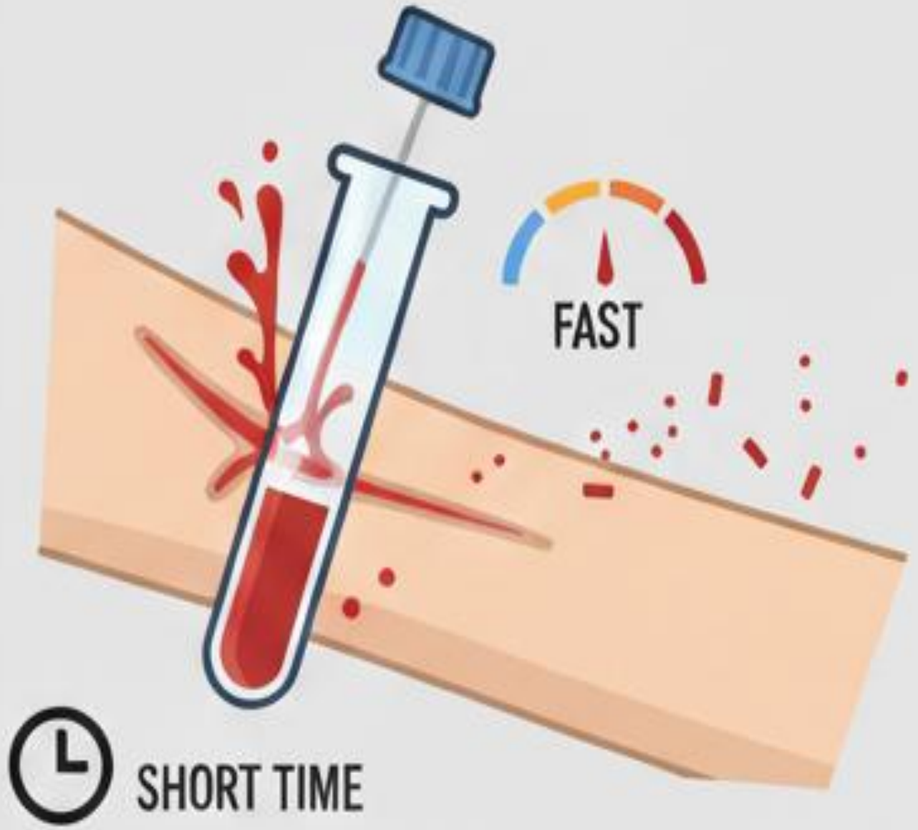
1. Tube does not contain an anticoagulant
2. It does contain a clot activator and serum separator gel
3. During centrifugation, this gel moves up to the interface between serum and clot, forming a stable barrier and separating the serum from the cells
4. This tube is used for collecting serum.
5. Most common tubes used in chemistry tests, such as the Liver Panel, Lipid Panel

CAUSES OF HEMOLYSIS DURING BLOOD COLLECTION

1. FORCING BLOOD THROUGH NEEDLE

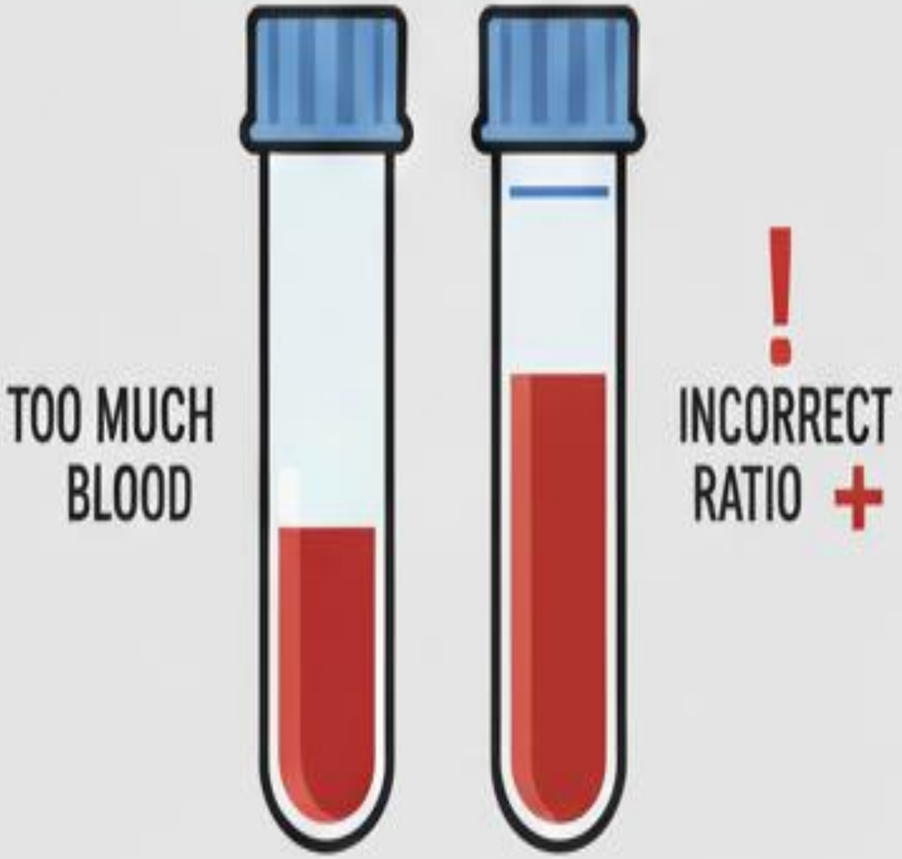


2. DRAWING BLOOD TOO QUICKLY



3. VIGURIUS SHAKING OR MIXING MIXING

4. IMPROPER TUBE FILLING

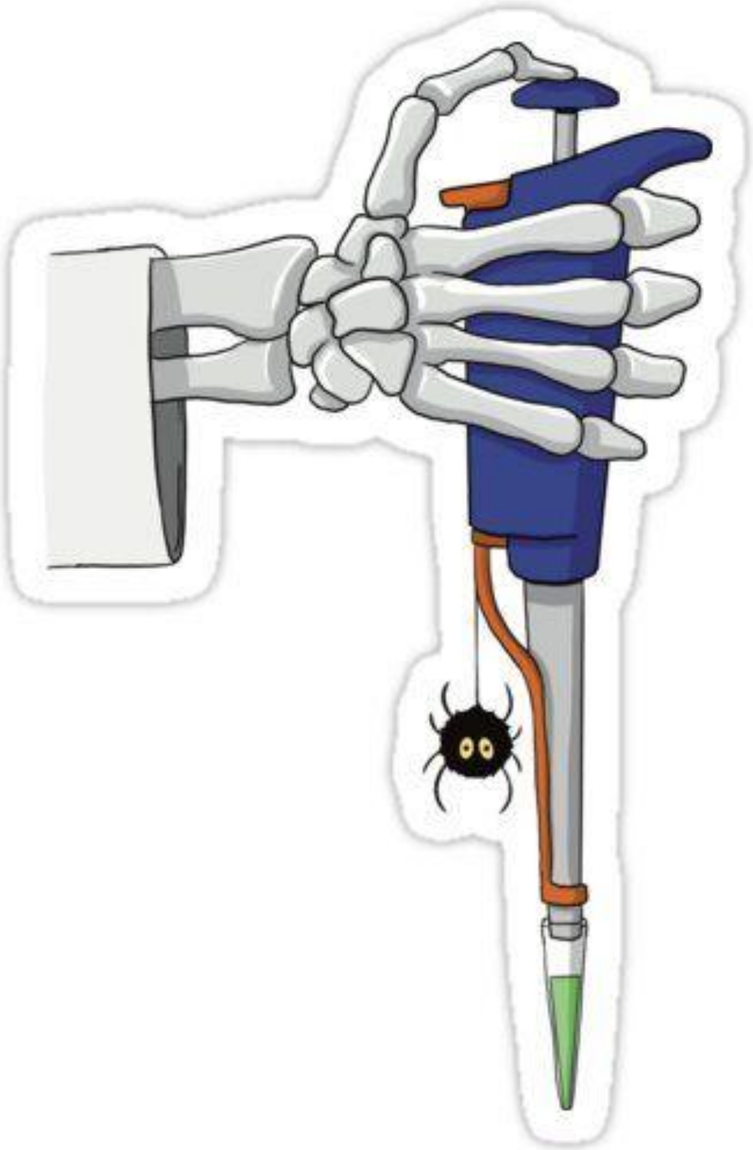


Case Study

- A 54-year-old man was admitted to the hospital after taking his breakfast for preoperative evaluation and elective knee surgery. On the morning of surgery, the patient was awakened by the technician, who drew blood for basic laboratory tests, including calcium, CBC, and Blood group, as well as PTT. The technician drew a blood sample using a syringe and tourniquet and discharged it in a plain tube
- **What important question must be asked by the technician before sample collection**
- **Which tubes must be used?**
- **What error did the technician make?**

Case study 2

- A blood sample had been taken from a 44-year-old woman in the medical ward, and the laboratory staff filled a container, forcing the blood through the needle with the needle attached to the syringe. The blood investigation result showed a falsely increased potassium level. Explain why.



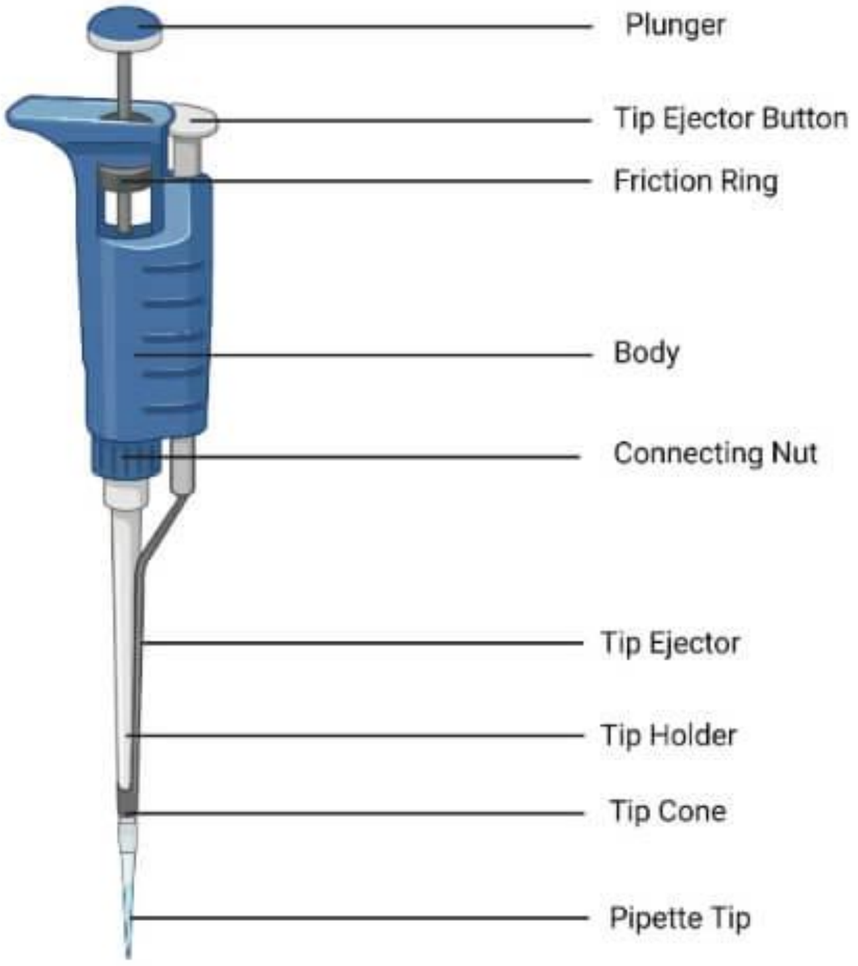
Micropipettes

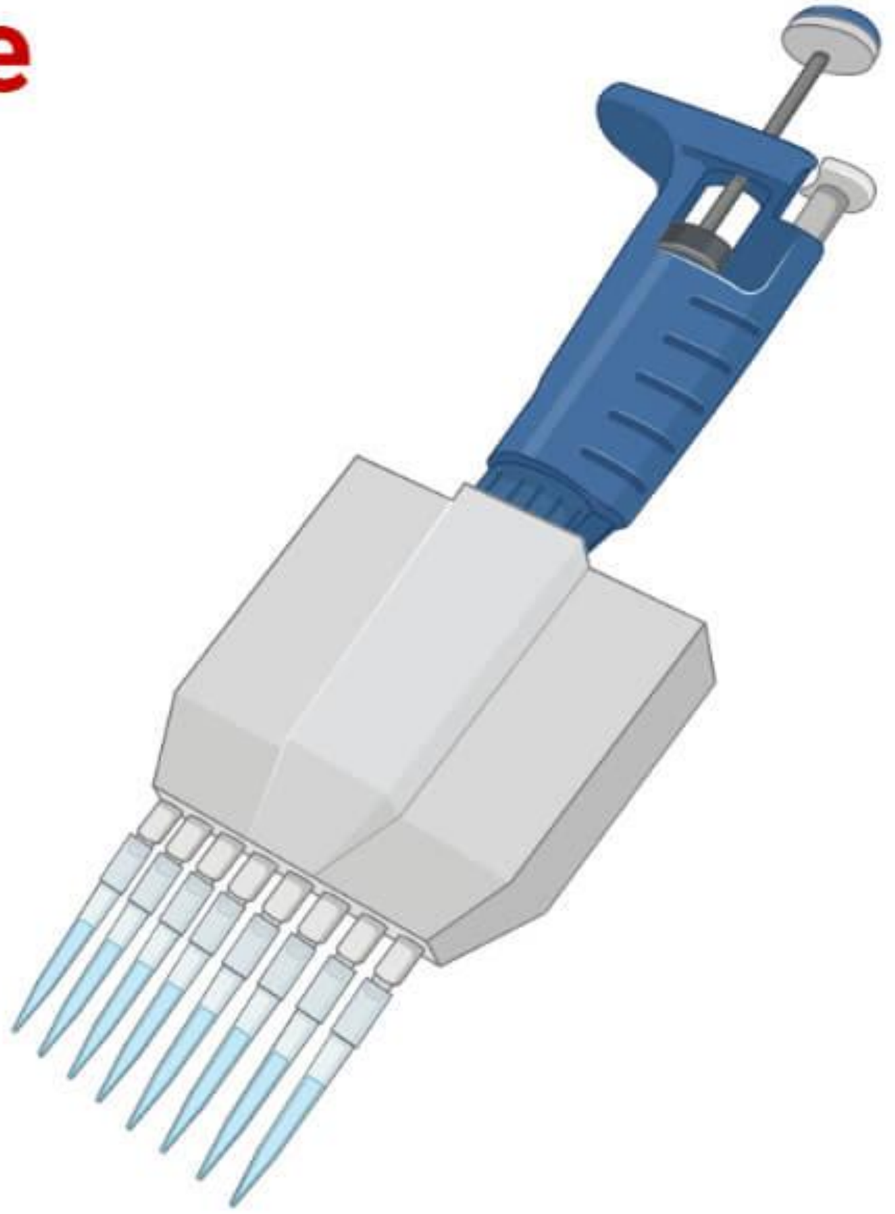
Micropipettes

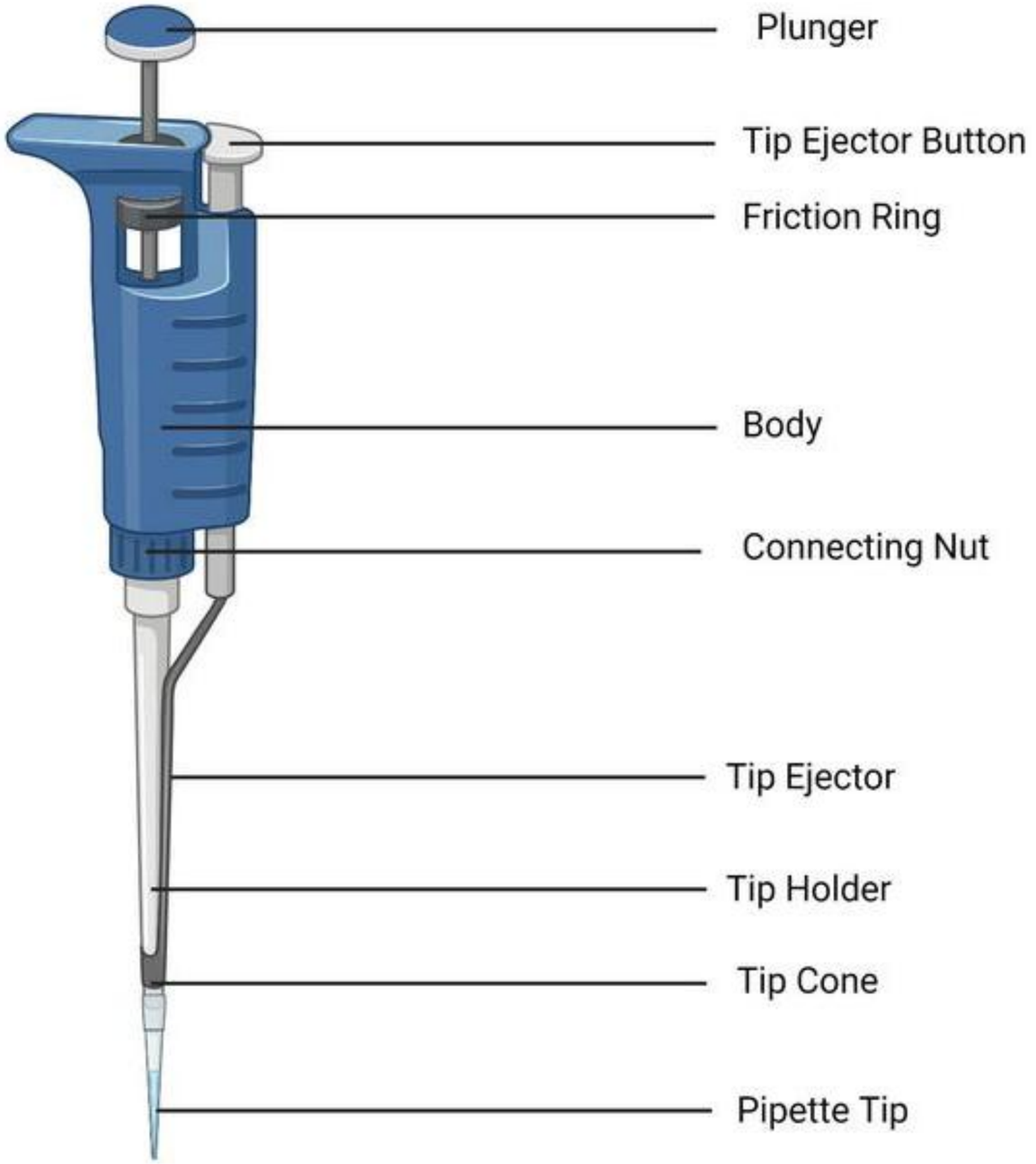
Micropipettes are precision instruments that are designed to accurately and precisely transfer volumes in the microliter range

1 microliter (abbreviated μL) = 10^{-3} milliliter (mL) = 10^{-6} liter (L)

Micropipette







Types of tips that are used in a chemistry lab

Type of Micropipette

1. **Fixed-volume micropipette:** fixed amount of liquid to be aspirated or dispensed

2. Variable volume micropipette: The amount of liquid to be aspirated or dispensed can be changed according to the need.

Applications of Micropipette

1. Chemical Laboratories
2. Pharmaceutical Laboratories
3. Molecular biology Laboratories

Steps to use a pipette

1. Setting the Volume
2. Attached The Pipette Tip
3. Depressing the plunger

Note: The first stop is used to fill the micropipette tip, and the second stop is used to dispense the contents of the tip

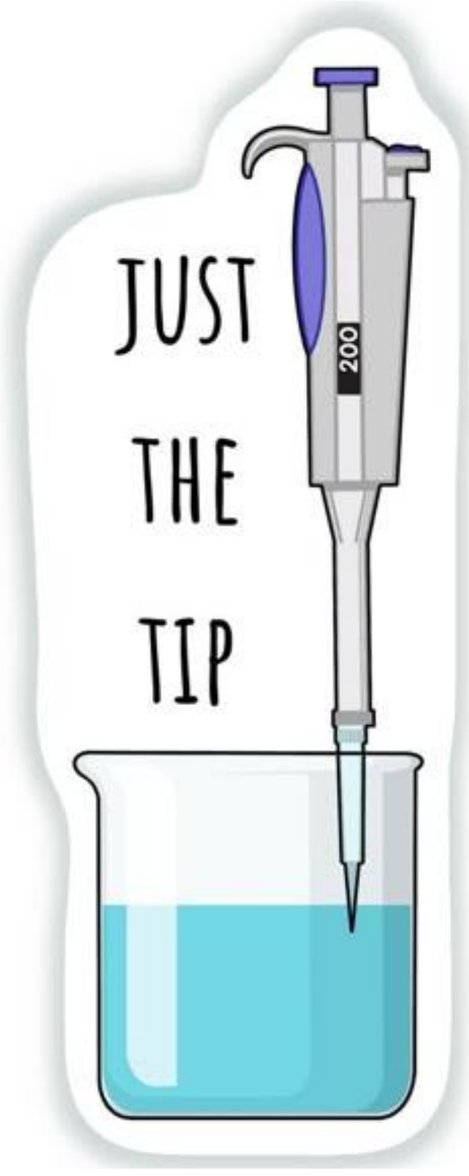
Start

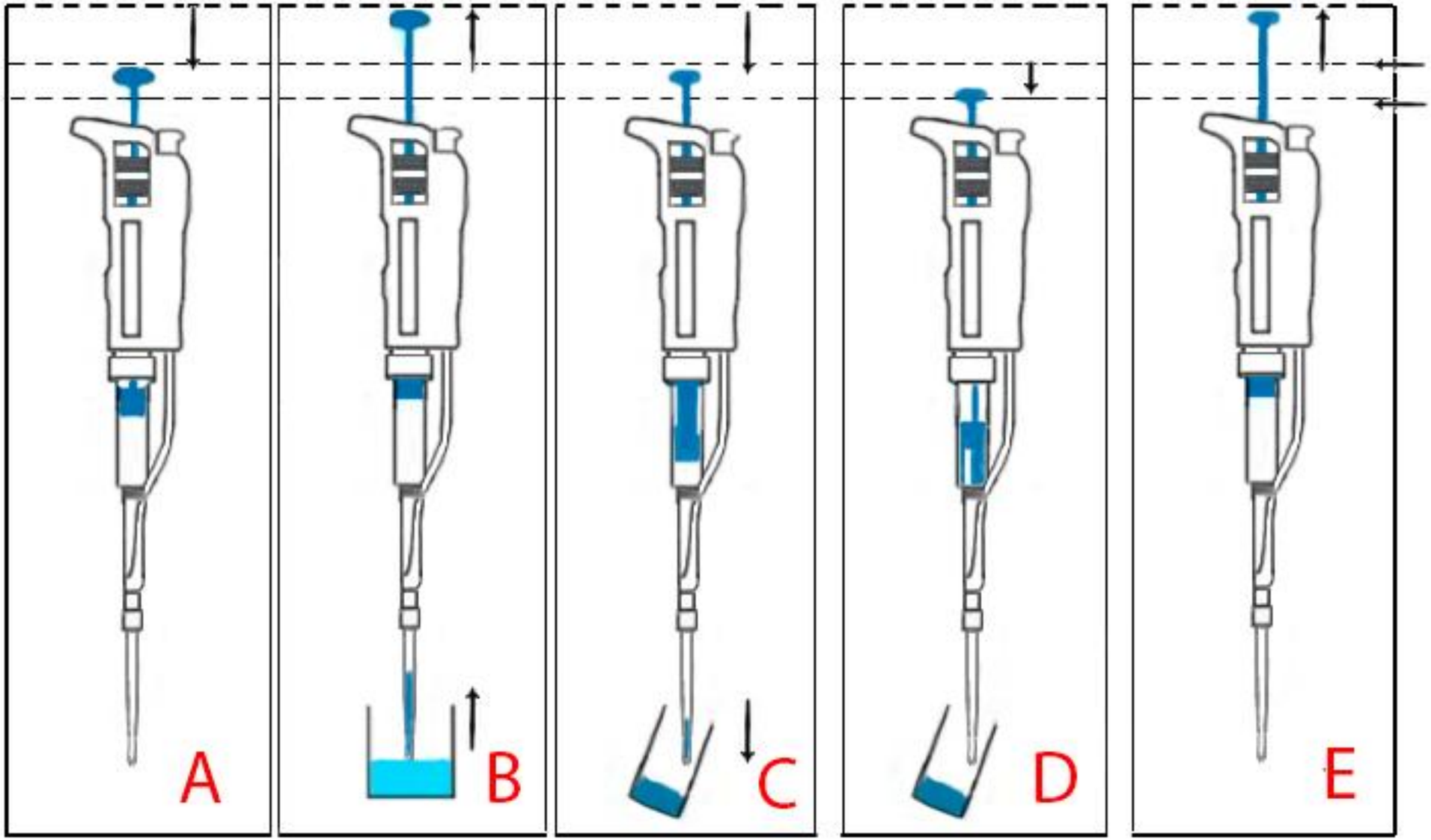
First stop

Second stop

Steps to use a pipette

1. Immerse Tip in Sample
2. Draw up the sample
3. Put the tip in the second tube
4. Depress the plunger to the second stop to dispense the sample
5. Withdraw the pipette from the tube
6. Discard the tip by pressing the tip ejector button

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Thank you for listening

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