



# Ensuring Accuracy: Best Practices for Patient Preparation and Sample Collection in Hematology, Cytopathology & Histopathology

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# **Blood SAMPLE COLLECTION & PROCESSING**

# PURPOSE FOR BLOOD COLLECTION...

- Biochemical  
eg. - S. electrolyte, S. proteins etc.
- Hematological  
eg. - CBC, Hb%, ESR etc.
- Serological  
eg. - Blood c/s etc.
- Genetic  
eg. – Cytogenetic and molecular genetic tests etc.

# STEPS OF BLOOD ANALYSIS...

Blood analysis is one of the most important diagnostic tools needed for clinician within healthcare & the steps are -

- 1) Collection.
- 2) Transport.
- 3) Preservation.
- 4) Processing.
- 5) Test procedure.
- 6) Report preparation and delivery.

## FOR COLLECTION...

- Verify the patient and exam request.
- Prepare for phlebotomy & it's the most important among them.
- Label sample tube with patient's ID.

- This procedure emphasizes the extreme importance of phlebotomy process.
- Studies show that the majority (68%) of laboratory errors occur in the pre-analytic stage, including phlebotomy process (WHO manual 2010)

# PROCEDURE FOR DRAWING BLOOD OR PHLEBOTOMY...

- Step 1. Identify and prepare the patient.
- Step 2. Assemble equipment.
- Step 3. Time of Collection.
- Step 4. Select the site.
- Step 5. Perform hand hygiene and put on gloves.
- Step 6. Disinfect the entry site.
- Step 7. Taking of blood.
- Step 8. Drawing samples in the correct order.
- Step 9. Cleaning contaminated surfaces and complete patient procedure.

## Step : 1

- Identify & prepare the patient.

## ERRORS...

- ✓ We must avoid any error in Patient Identification.
- ✓ Patient Variables also affect Blood Specimens due to lack of specific instructions to the patient to avoid those. Such as -
  - **Diet** : Consider fasting requirements or specific dietary restrictions.
  - **Obesity** : Obesity can influence blood sample collection and analysis.
  - **Therapeutic drug monitoring** :  
Example - If patient is on heparin therapy & patient give blood sample after taking heparin, forgot to give blood before taking it.

- **Allergies** : Alcohol or iodine sensitivity used for venipuncture site cleaning. Consider alternative cleaning agents if necessary.
- **Drug Interactions** : Certain medications can alter blood test results. So avoid on going drug prescription for 8 hours.
- **Physical Activity/ Exercise** : Avoid strenuous activity within 20 mins before blood collection. Neutrophil level increased after physical activity.
- **Smoking** : Nicotine and other components in tobacco can impact blood parameters.
- **Stress** : Emotional stress may affect blood test outcomes.



## Step : 2

### • Assemble equipment

#### Phlebotomy tray

- Disposable syringes and sterile needles (23 gauge needle).
- Tourniquet.
- Container for blood collection (plain tube and anticoagulants).
- Rectified spirit (70% alcohol or 0.5% chlorohexidine).
- Swab/cotton/pad.
- Adhesive dressing.
- Racks to hold specimen upright.
- Requisition form .
- Disposal container.



## Different types of needle for phlebotomy...

- Straight needle - Commonly used needle for blood drawing.
- Butterfly needle.
- Nowadays, blood is collected using a vacutainer syringe. The syringe is connected to a collection tube, and blood flows into the tube due to the negative pressure created by it.



## Anticoagulants...

- EDTA
- 3.8% Tri sodium citrate
- Heparin
- Sodium fluoride
- Double oxalate or Paul Heller's mixture
- Potassium oxalate
- Acid citrate dextrose
- Citrate phosphate dextrose



Plain tube/  
clot  
activator  
tube



EDTA  
tube(5.4mg)



Sodium  
fluoride(3mg)  
& EDTA  
(2.6mg)contai  
ning tube







Tri-sodium  
citrate(3.2  
%)containi  
ng tube



ESR tube: containing 3.8%  
tri-sodium citrate

# Anticoagulants...

Name	Mechanism of action	Concentration used	Use
 EDTA (ethylene diamine tetra acetic acid)	Prevents blood clotting by removing calcium ion by chelation	1.2 mg/ml of blood	<ul style="list-style-type: none"> <li>All hematological procedure (CBC, Hb, RBC, TC, DC, PCV, RBC indices, PBF, Platelet count, Reticulocyte count, HbA1C,</li> <li>Body fluid.</li> </ul>
 3.8% Trisodium citrate		ESR: 1.6ml blood in 0.4ml TSC Coagulation studies: 9 ml blood in 1 ml TSC	<ul style="list-style-type: none"> <li>ESR estimation</li> <li>Coagulation studies</li> </ul>
 Heparin	inactivating thrombin and preventing the conversion of fibrinogen to fibrin.	0.1-0.2mg/ml of blood	<ul style="list-style-type: none"> <li>ABG,</li> <li>Osmotic fragility of RBC, some biochemical and emergency tests like Troponin-I</li> </ul>
 Sodium fluoride	Inhibit glucose metabolism by inhibiting enolase enzyme (inhibit glycolysis)		Blood glucose estimation

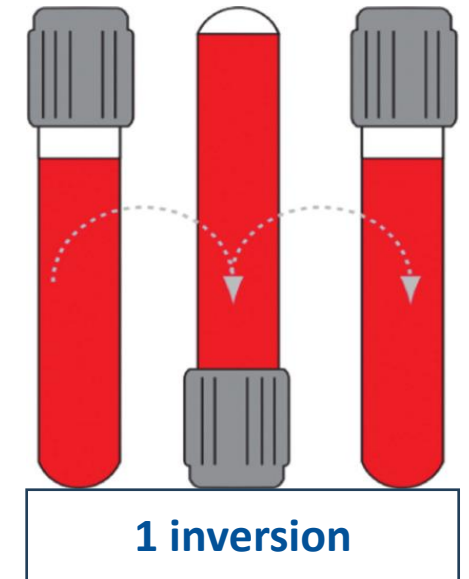
# ERRORS...

- ❖ **Hygiene** : Lack of strict hygiene measure of the equipment can lead to septic condition.
- ❖ **Tube Type** :
  - Appropriate tubes for specific tests.  
Example - light blue top tube for Coagulation Studies.
  - Incomplete filling results in specimen dilution and erroneous Prothrombin and APTT test results.
- ❖ **Anticoagulant Levels** :
  - Inappropriate anticoagulant in collection tubes or Insufficient or excessive anticoagulant levels can affect test results.
- ❖ After cleaning the site with 70% alcohol, we should allow it to dry. Otherwise it could contaminate the sample and causes hemolysis.

## ❖ **Mixing :**

Mix blood samples adequately after collection.

Inadequate or excessive mixing can impact accuracy.



## ❖ **Prolonged Tourniquet Application :**

Prolonged use can cause hematoma and hemoconcentration.

Also there are evidences of increasing K<sup>+</sup>, total protein, lactic acid due to prolong torniquetion > 1 min.

❖ Blood shouldn't be drawn very rapidly (to avoid hemolysis) or too slowly (to avoid some coagulation).

## Step : 3

- Time of collection.

**ERRORS...**

- **Diurnal Variation :**

Blood test results vary throughout the day (due to natural biological rhythms).

Specific instruction to collect blood early in the morning should be given. Ill timing during collection may bring wrong result.

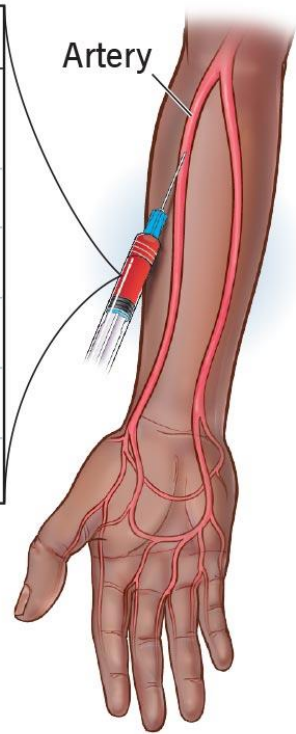
Example - In the evening cortisol level is high.

- Some tests should perform within 15 minutes of sample collection. eg. - Arterial Blood Gas analysis (ABG).

## ABG is an urgent test...

### Arterial Blood Gas (ABG)

ABG	Normal range
O <sub>2</sub> CT	15-23% per 100 mL of blood
pH	7.35-7.45
PaCO <sub>2</sub>	35-45 mmHg
PaO <sub>2</sub>	80-100 mmHg
HCO <sub>3</sub>	22-26 mEq/L
O <sub>2</sub> Sat	95-100%



- There is a significant decrease in both PaO<sub>2</sub> and PaCO<sub>2</sub>, along with changes in pH, within 15 minutes.
- Sodium and potassium levels show changes within 60 minutes when kept at room temperature.

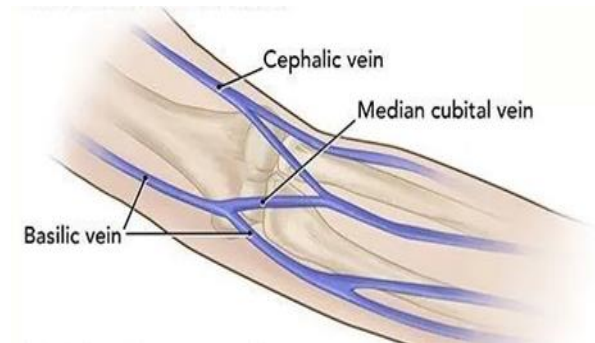
*Therefore, it's crucial to perform Arterial Blood Gas (ABG) analysis within 15 minutes of sample collection.*



## Step : 4

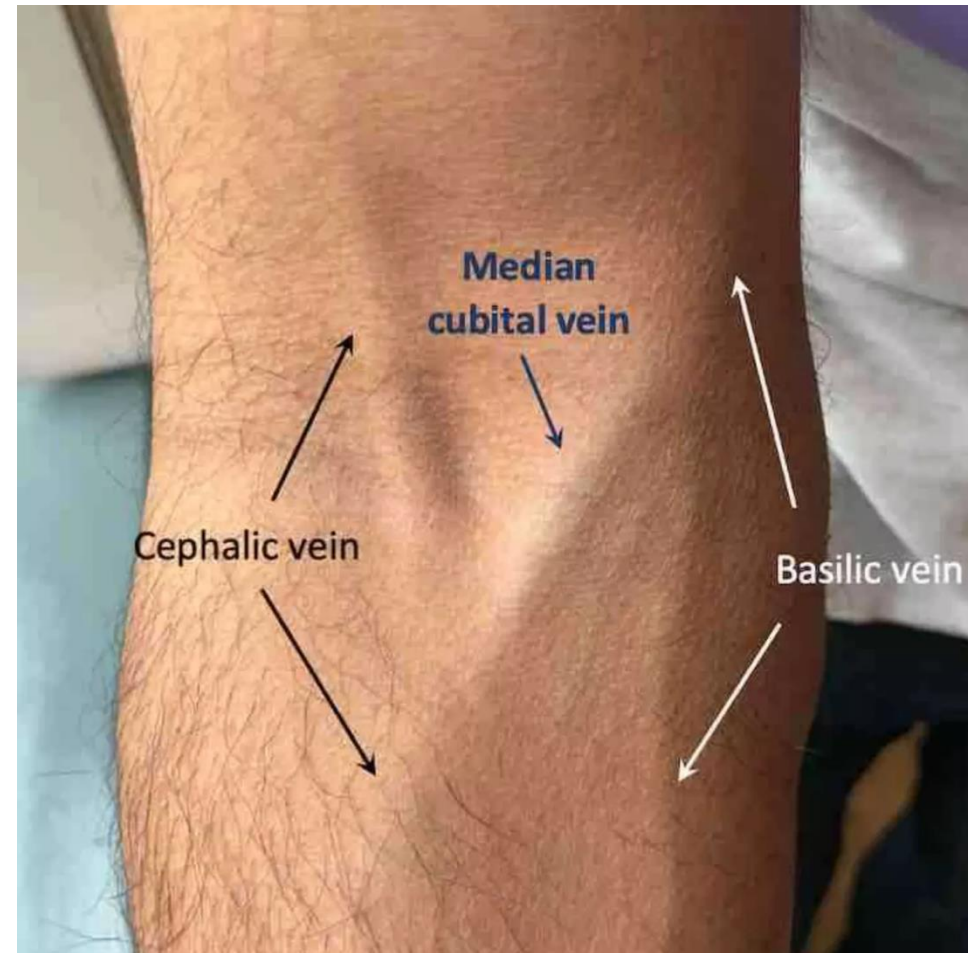
### • Select the site.

- The word "Phlebotomy" derives from the Greek "phleps," meaning "vein," and "tomia," meaning "cutting."
- the Median cubital vein is usually the vein of choice for phlebotomy: It is typically more stable (less likely to roll), it lies more superficially, and the skin overlying it is less sensitive than the skin overlying the other veins.
- The other veins we use for phlebotomy are cephalic vein and basilic vein.



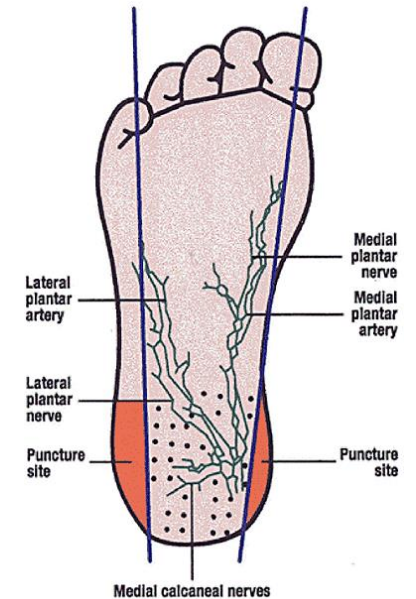
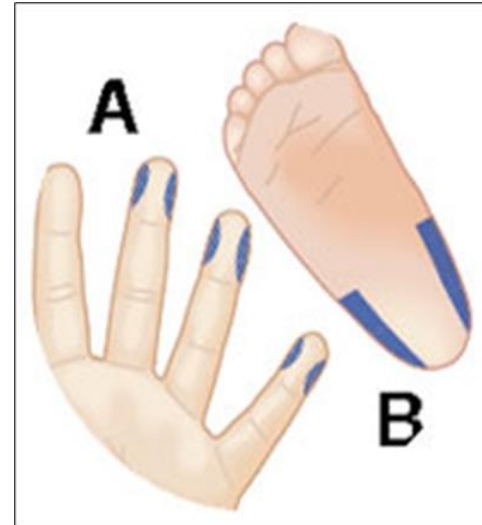
# Venous blood collection: Venipuncture...

- Venous blood is collected by appropriately trained laboratory staff with strict aseptic precautions.
- Vein: Median cubital vein



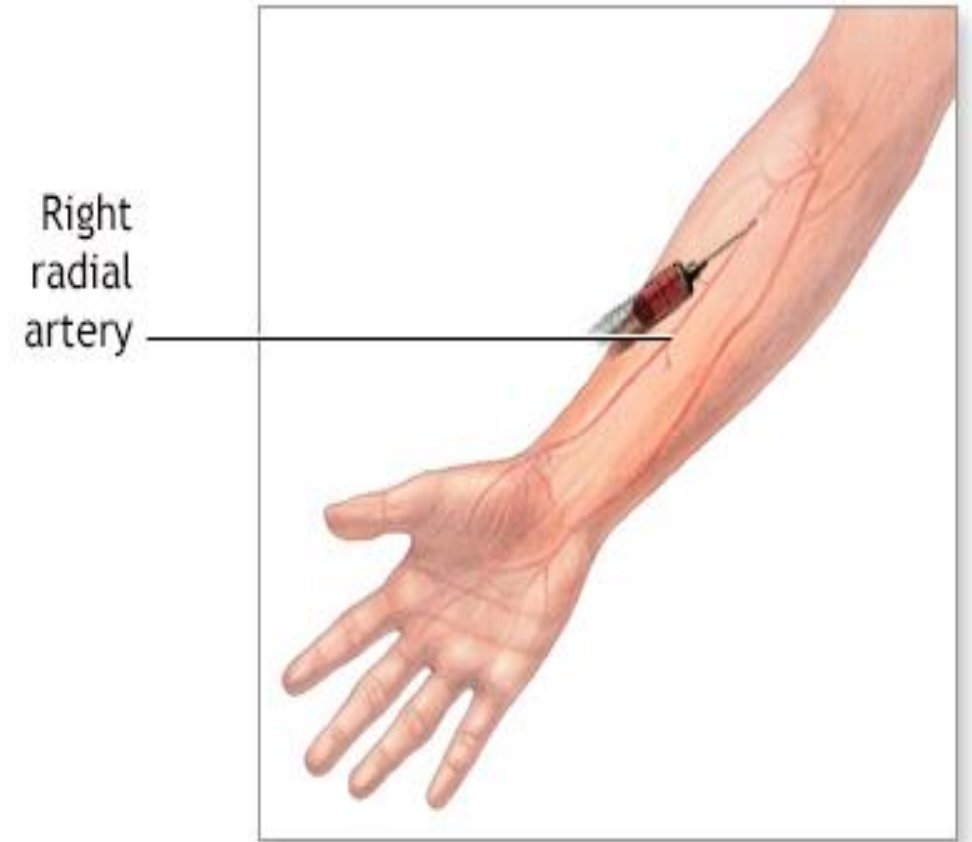
# Capillary blood collection...

- Capillary blood can be collected with a lancet or needle.
- For older children and adults:
  - **distal digit** of the 3rd or 4th finger on its palmar surface, approximately **3-5mm lateral from the nail bed**.
- For infants:
  - **deep puncture** in the plantar area of the heel.



# Arterial blood collection...

- Arterial blood required rarely.
- Site: Femoral, radial or brachial artery.
- Use: Estimation of blood gases.



# Difference between capillary and venous blood

Parameter	Capillary Blood	Venous Blood
PCV, Hb%, RBC count	Higher	Lower
TC, Neutrophil count	Higher by about 8%	Lower
Monocyte count	Higher by about 12% (children up to 100%)	Lower
Platelet count	Lower	Higher on average by about 9% (may be up to 32%)
Glucose levels (prandial state)	20-25% higher	Lower
Glucose levels (fasting state)	2-5 mg/dl higher	Lower
Potassium, Calcium, Total protein	Lower	Higher

# Blood specimen types with their common uses...

Specimen type	Method of collection	Uses
Capillary blood	Skin puncture in finger tips, ear lobe or heels	<ul style="list-style-type: none"><li>• Useful when only a small sample is needed.</li><li>• In case of infant or child.</li><li>• In case of elderly patient with fragile veins.</li><li>• Severe burned patients.</li><li>• Monitor anemia during pregnancy in distant laboratories.</li><li>• Commonly done:<ul style="list-style-type: none"><li>➤ Thick and thin films</li><li>➤ WBC total count</li><li>➤ Hb estimation</li><li>➤ Bleeding time and clotting time.</li></ul></li></ul>
Venous blood	Venipuncture	All laboratory procedures.
Arterial blood	Direct puncture on artery	Arterial blood gas analysis.

## ERRORS...

- **Avoid Sites with Intravenous fluid infusion:**

- ✓ Choose an alternative arm or draw below if Intravenous fluid is transfused.

*This prevents contamination or dilution of the blood sample due to the IV fluids.*

- ✓ There is also risk of Infection.

- **Edematous areas :** Avoid due to fluid accumulation.

*It may lead to specimen contamination or dilution.*

- **Obstructed, hardened, scarred veins:** *Not suitable for collection.*

Step :4

• Perform hand hygiene and put on gloves





Step :5

- Disinfect the entry site



Step :6

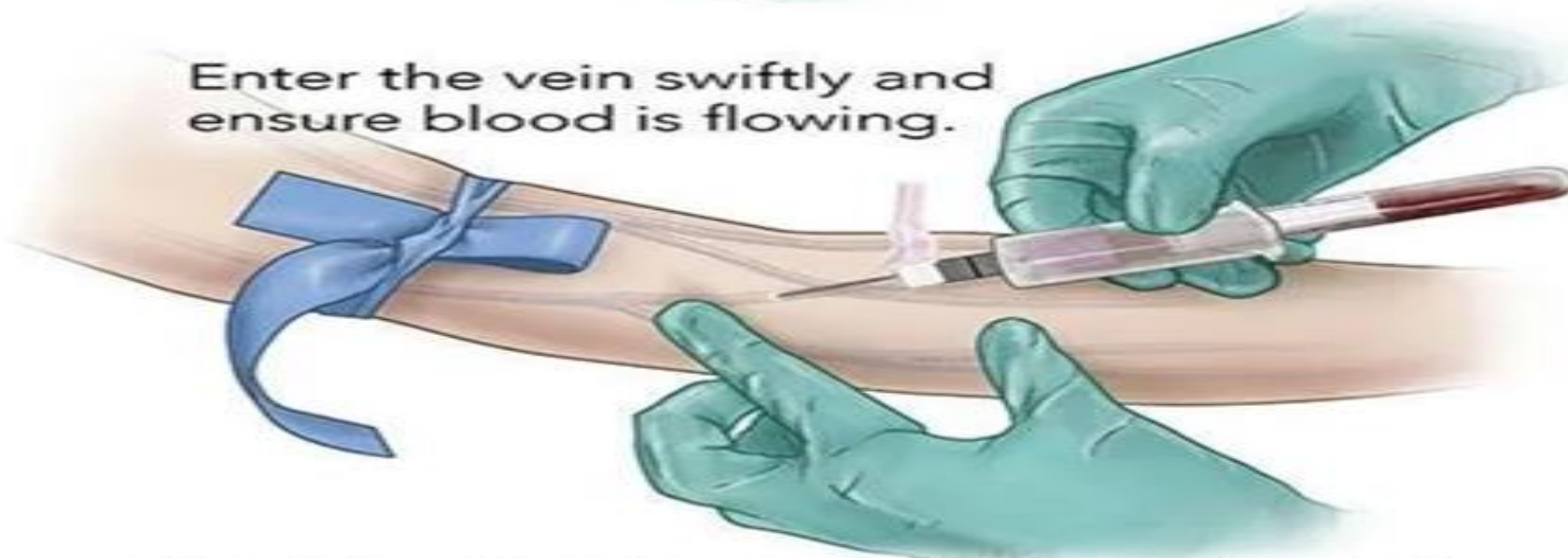
• Drawing samples in the correct order

Anchor the selected vein with thumb and index finger.



Position the needle at a 30° angle.

Enter the vein swiftly and ensure blood is flowing.

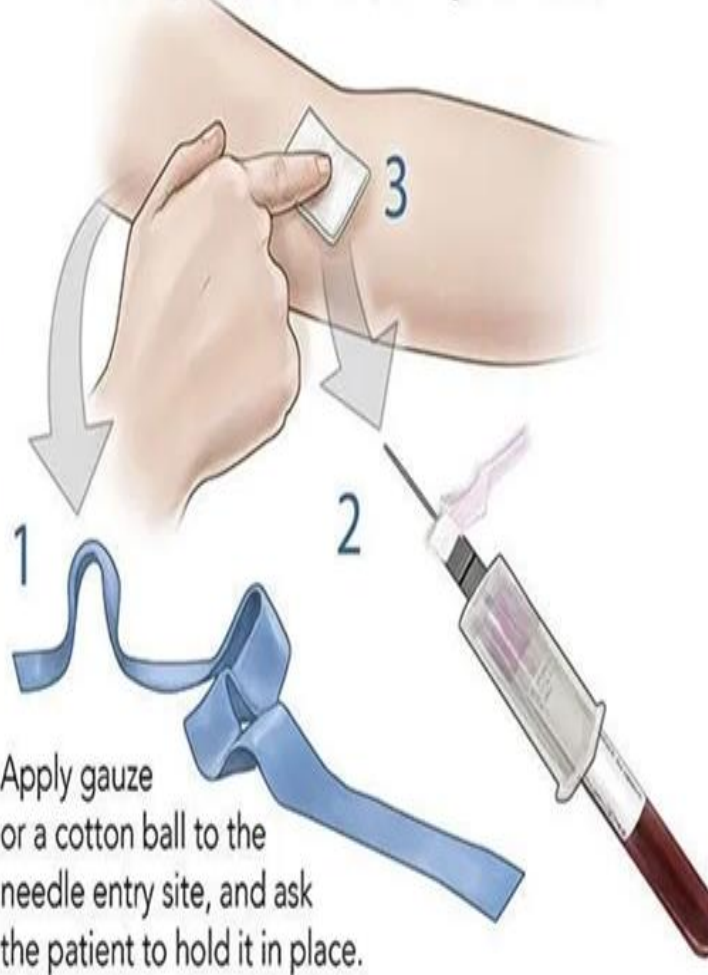


After blood has been collected, release the tourniquet *before* withdrawing the needle.

## Step :7

# • Cleaning contaminated surfaces and complete patient procedure

After blood has been collected, release the tourniquet before withdrawing the needle.



Remove and immediately invert the tube 8 to 10 times to mix the sample with the tube additives.



Label the tube for transport to the lab, indicating:

- Patient's full name
- Patient ID
- Birth date
- Date of sample.



# ERRORS...

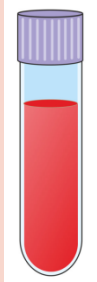
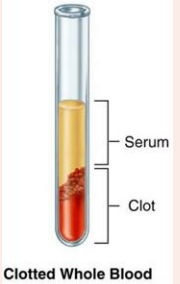
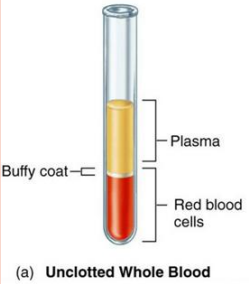
## ❖ **Technical error :**

- ✓ Due to improper preparation of venipuncture site.
- ✓ Faulty technique.
- ✓ Sampling error.
- ✓ Improper sealing.
- ✓ Inappropriate discarding of needle.

# ERRORS DURING TRANSPORT...

- ❖ Must be transported at the appropriate temperature for the required test (On ice - ABG, Ammonia).
- ❖ **Transportation faults.**
  - Some specimens must be transported immediately after collection, for example Arterial Blood Gases (within 15 mins).
  - should be centrifuged and separated within two hours.

# Blood components...

Blood component		Use
Whole blood 	Blood sample (venous, arterial, or capillary) where the cellular and extracellular properties closely resemble the in vivo state.	<ul style="list-style-type: none"> <li>• CBC</li> <li>• ABG analysis</li> <li>• Blood glucose and HbA1C estimation.</li> <li>• ESR estimation.</li> </ul>
Serum 	Undiluted, extracellular portion of blood after complete blood coagulation	<ul style="list-style-type: none"> <li>• Biochemical Tests</li> <li>• Tumor Marker and Other Immunological Tests</li> <li>• Hormone Study</li> </ul>
Plasma 	The virtually cell free supernatant of blood containing anticoagulants obtained after centrifugation.	<ul style="list-style-type: none"> <li>• Coagulation profile</li> <li>• Blood glucose estimation</li> </ul>

# Storage of blood sample...

## Whole Blood:

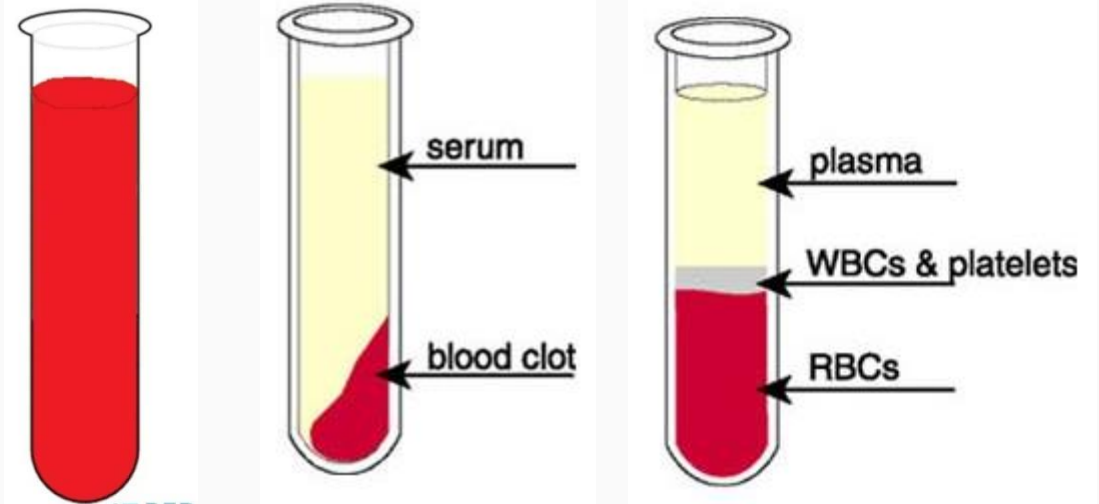
- 4-8 hours at room temperature.
- 24 hours at 4-8°C.

## Coagulation Tests (Plasma):

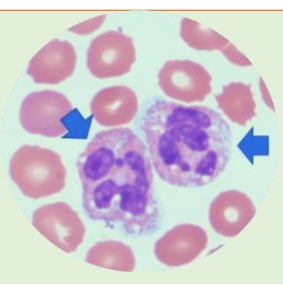
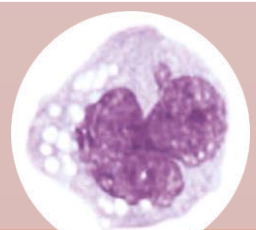
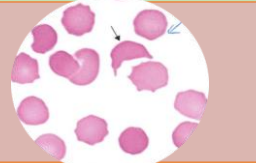
- Within 2 hours at room temperature.
- Within 4 hours at 4°C.
- Up to 2 weeks at -20°C.
- Up to 6 months at -70°C.

## Serum or Plasma Stability:

- Remains stable for 5 hours after collection
- Can be stored at -20°C for > 2 - 3 weeks.



# EFFECT OF STORAGE...

Neutrophils	<ul style="list-style-type: none"><li>• Nuclear lobe separation</li><li>• Irregular cytoplasmic margin</li><li>• Appearance of cytoplasmic vacuoles</li><li>• Degeneration</li></ul>	 A circular microscopic image showing a group of neutrophils. Two neutrophils in the center have nuclei with distinct lobes and gaps between them, indicated by blue arrows. The cytoplasm of these cells contains small, clear vacuoles.
Monocytes	<ul style="list-style-type: none"><li>• Small vacuoles in the cytoplasm</li><li>• Irregular lobulation of the nucleus (segmentation)</li><li>• Cellular disintegration</li></ul>	 A circular microscopic image of a single monocyte. The nucleus is large and has an irregular, segmented appearance. The cytoplasm is filled with numerous small, clear vacuoles.
Lymphocytes	<ul style="list-style-type: none"><li>• Small vacuoles in the cytoplasm</li><li>• Nucleus stain more homogeneously</li><li>• Nucleus undergoes budding to give rise to nuclei with 2-3 lobes</li></ul>	
Red blood cells	<ul style="list-style-type: none"><li>• Crenation and sheering occurs.</li></ul>	 A circular microscopic image showing a group of red blood cells. Some cells are shrunken and have a spiky, crenated appearance, while others are fragmented, as indicated by blue arrows.
Platelets	<ul style="list-style-type: none"><li>• Disintegration</li></ul>	
Others	<ul style="list-style-type: none"><li>• With time, PCV, MCV and osmotic fragility increases</li><li>• ESR decreases</li></ul>	



# ERRORS DURING SPECIMEN HANDLING...

- Ensure appropriate handling procedures.
- Avoid contamination or mishandling.

## ❖ **Inappropriate Storage:**

Store specimens correctly.

Follow temperature requirements for specific tests (e.g., ABGs, Ammonia).

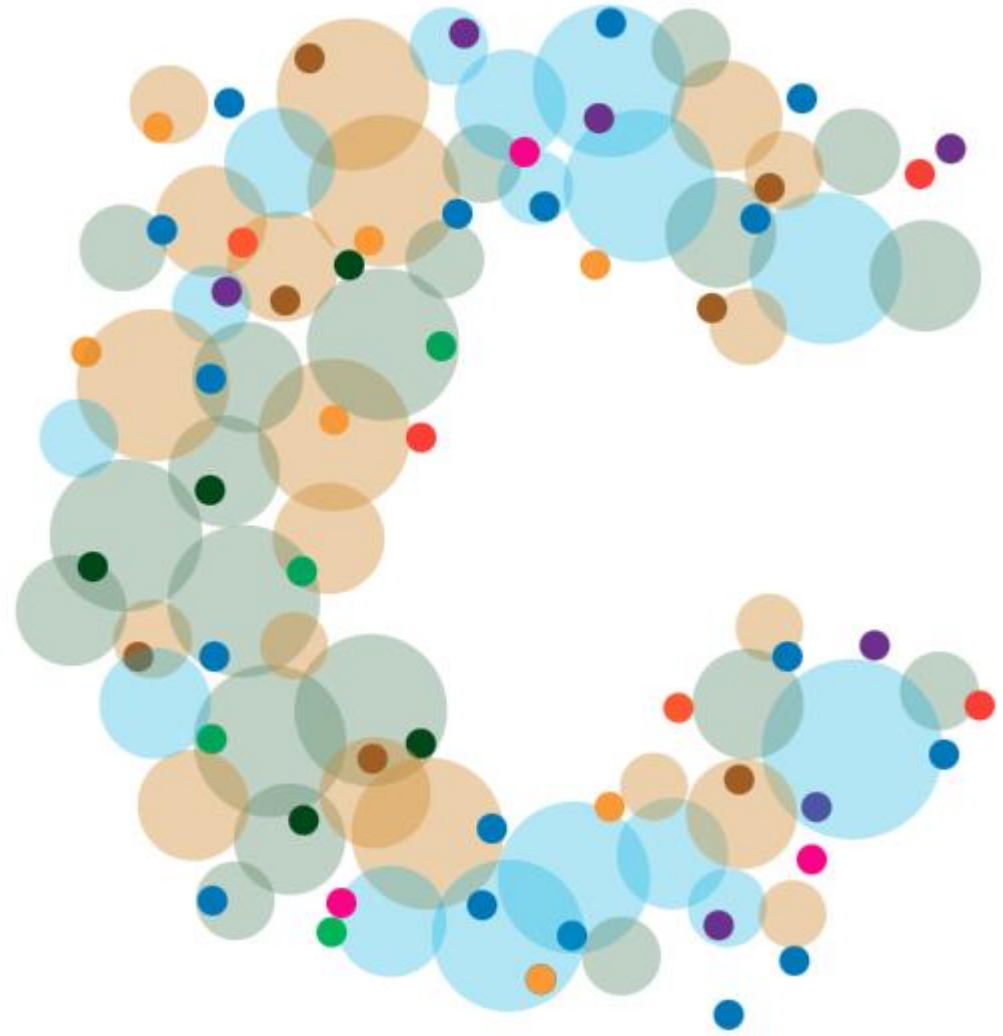
## ❖ **Timely Separation:**

Delay in separation of serum or plasma from whole blood may alter result.

Centrifuge and separate within two hours to maintain sample integrity.

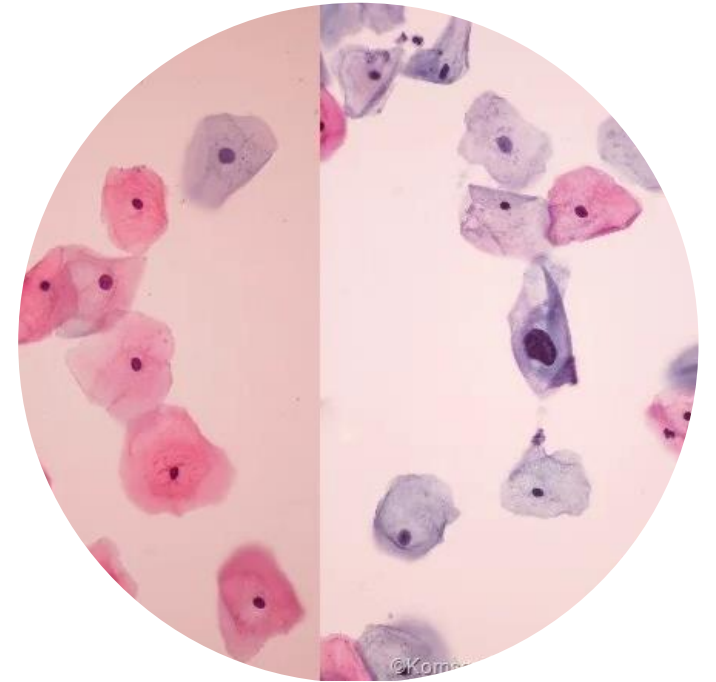
# Cytopathology sample collection

Dr. Andrila Banik Shaily, Lecturer  
Department of Pathology



# Cytopathology

Diagnostic techniques that are used to **examine cells** from various body sites to determine the **cause** or **nature of disease**.



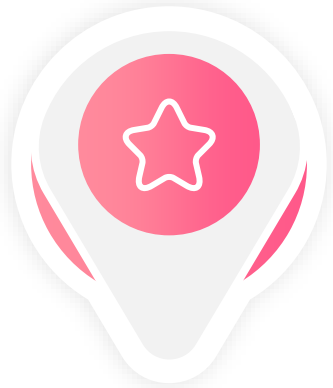
# Sampling techniques

Diagnostic cytology is based on 4 basic sampling techniques:

## Abrasive cytology

Collection of cells removed by brushing or similar abrasive techniques

## Intraoperative cytology



## Exfoliative cytology

Collection of exfoliated cells



## Fine Needle Aspiration Cytology

Removal of cells from palpable or deeply seated lesions by means of a needle



# Exfoliative cytology

Based on spontaneous shedding of cells derived from the lining of an organ into a cavity

Examples:

✓ sputum

✓ urine

✓ body fluids - effusions

- Other fluids e.g. CSF, Synovial

fluid etc



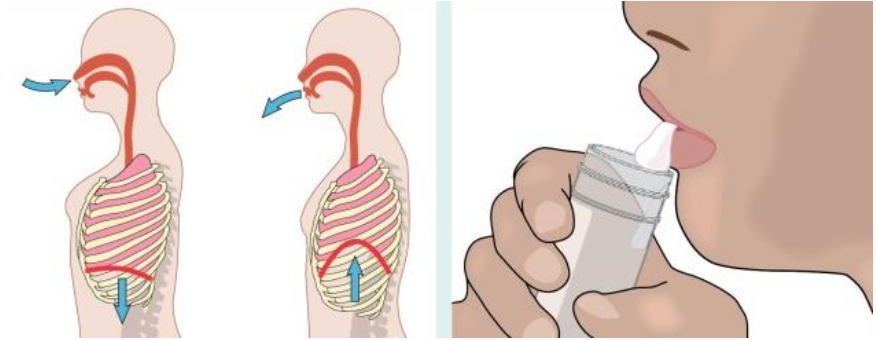
Exfoliative cytology

# Sputum

Instructions for collecting a sputum specimen:

1. Morning specimen is required

i.e., Before breakfast after rinsing mouth with water (avoid mouthwash).



- ✓ Rinsing with water will reduce contamination by saliva as saliva is of no diagnostic value.
- ✓ Mouthwash may kill the bacteria.

## Exfoliative cyto.. Sputum

2. Cough deeply & expectorate directly into containers with Saccomanno fixative and gently swirl to mix the specimen.

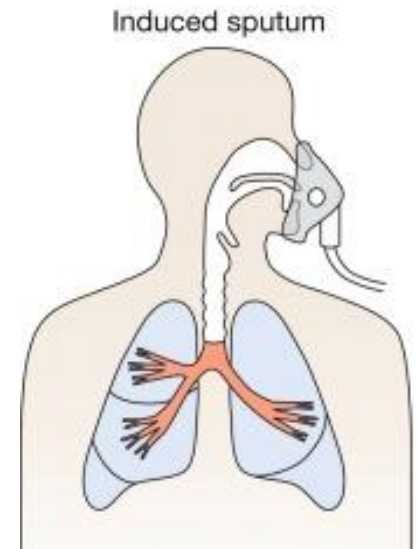
This special fixative will keep the cells well-preserved .

3. Do not use 95% ethanol.

Higher concentration of ethanol binds with proteinaceous material that form a hard lump, which makes it difficult to make thin-layered slides.

## Exfoliative cyto.. Sputum

4. Collect the specimen in the container, aiming for at least 1 teaspoon of sputum.
5. For better results, submit specimens on 3 to 5 successive mornings.
6. If this method fails, consider an induced sputum or tracheal aspirate.





Exfoliative cyto..

# Urine

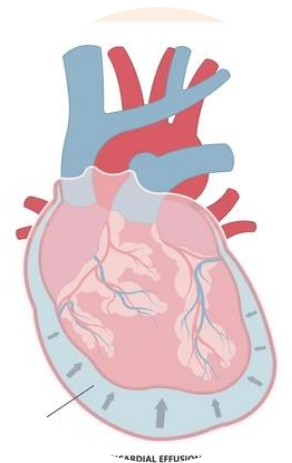
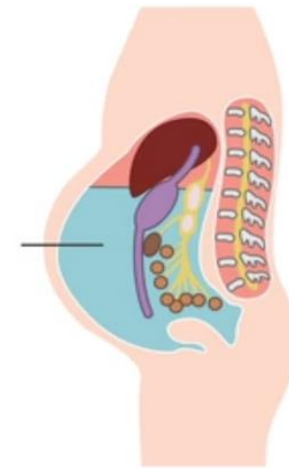
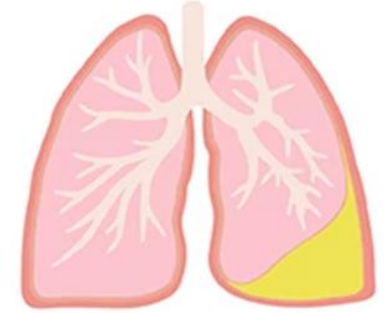
- Increase fluid intake of the patient before collecting the specimen.
- Avoid collecting samples during menstruation.
- Place the specimen in a plastic container with Saccomanno fixative.
- Label the container (not the lid) with the patient's name, age, and specify whether it's a voided or catheterized specimen.



# Exfoliative cyto..

## Body fluid

- ❑ Body cavity fluids are commonly evaluated for the presence of malignant cells from metastatic disease.
- ❑ These fluids include
  - effusion: pleural, peritoneal, pericardial
  - other fluid; CSF, synovial etc.
- ❑ No fixative should be added to body fluid specimens.  
(heparin may be added to prevent clotting)



## Exfoliative cyto.. Body fluid...

- A minimum of 10 mL of specimen is recommended; 50–100 mL is optimal for cytologic evaluation.

volume < 50 mL may not be optimal to exclude malignancy

- CSF cytopathology - volume not < 1-2 mL
- Specimens are collected in clean containers and with proper labelling dispatched immediately to the cytology laboratory.



## Green Life Medical College & Hospital

**Name:** ABC.....

**Age/Sex:** 50/female

**Hospital registration number:** 123456

**Procedure:** Left paracentesis

**Sample:** Ascitic fluid

**Amount sent:** 40mL

**Date and time of collection:** April 17, 2024. 9 AM

**Anticoagulant used or not, including type:** Heparin

**Clinical symptoms/history:** Weight loss and abdominal distension

**Clinical diagnosis:** Tuberculosis

**Imaging findings:** Left Adnexal mass

**Endoscopic findings, if any:**

**Any other relevant investigations:** CA-125 raised

**Relevant past history, if any:** None

**Investigation required:** Evaluation for Malignant cells

# Sample requisition form (Fluid cytology)

# Abrasive cytology

Cells are obtained directly from the surface of the target of interest.

Samples can be collected by-

- Scrapping
- Brushing
- Washing



# Abrasive cyt...

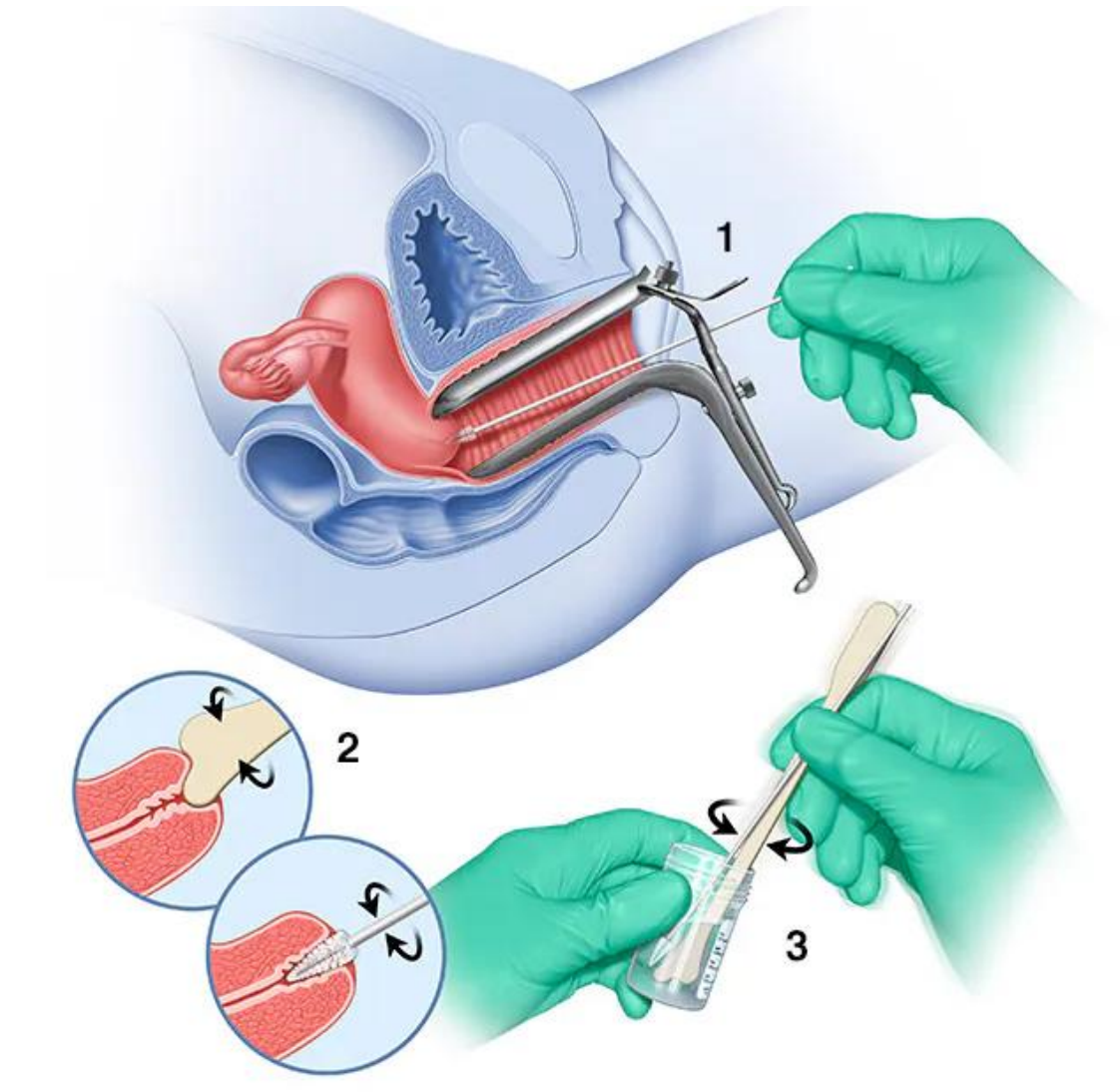
## Scrapping

### Paps smear

Provide-

- patient's age
- last menstrual period (LMP)
- any high-risk factor for cervical neoplasia

Such as “history of dysplasia” or “previous abnormal Pap result”



# Abrasive cyt...

## Paps sm..

### Time of collection of paps

- 2 weeks after the 1st day of LMP and if possible, not during menstruation.

- ✓ Heavy bleeding can obscure cellular detail.
- ✓ Menstrual smears are often covered with blood and inflammatory debris & contain few epithelial cells.

# Abrasive cyt...

## Paps sm..

### Patient Preparation

- Avoid douching & sexual intercourse for at least a day before the examination.
- Do not use intravaginal drugs or preparations for at least 1 week before the examination.

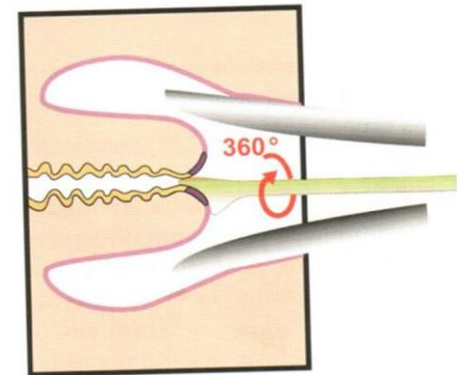
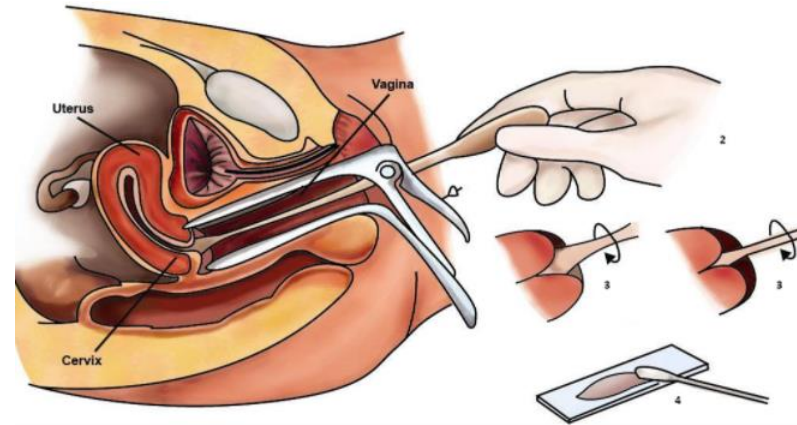
- ✓ Sexual intercourse may interfere with interpretation due to the presence of foams, jellies, sperm, or contamination of epithelium from the sexual partner.
- ✓ Both intercourse and douching cause exfoliation of epithelial cells and may result in scanty smears.

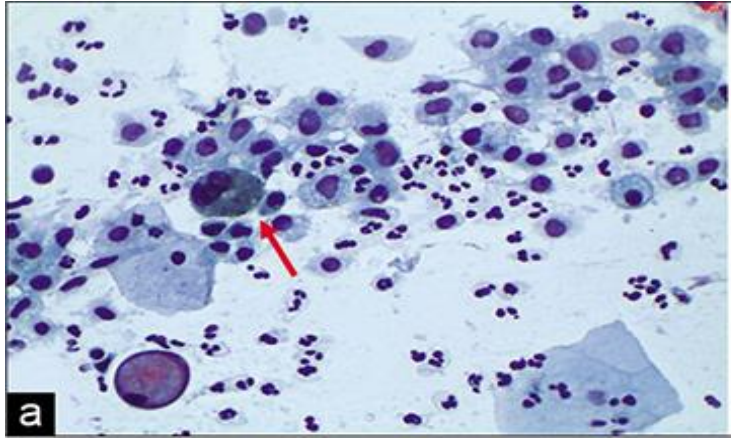


# Abrasive cyt... Paps sm..

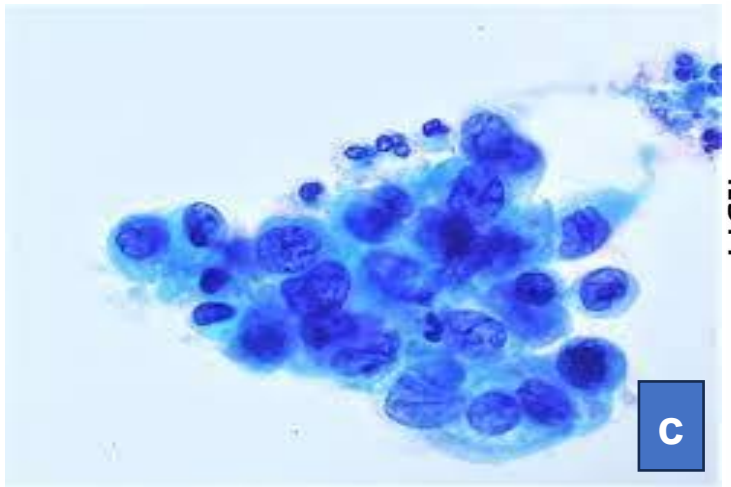
## Procedure to collect specimen

- Advice patient to void before the procedure.
- Lithotomy position on examining table
- Insertion of speculum without lubricant on it.
- Do not clean the cervix with saline prior to collecting the sample.
- Use spatula at the external os and rotate through 360° to collect sample.
- Collect the specimen from transition zone(squamous – columnar junction)





Acute cervicitis



High grade squamous intraepithelial lesion

# Abrasive cyt...

## Brushing Cytology

**Site:** Bronchial, esophageal, gastric, bile duct etc

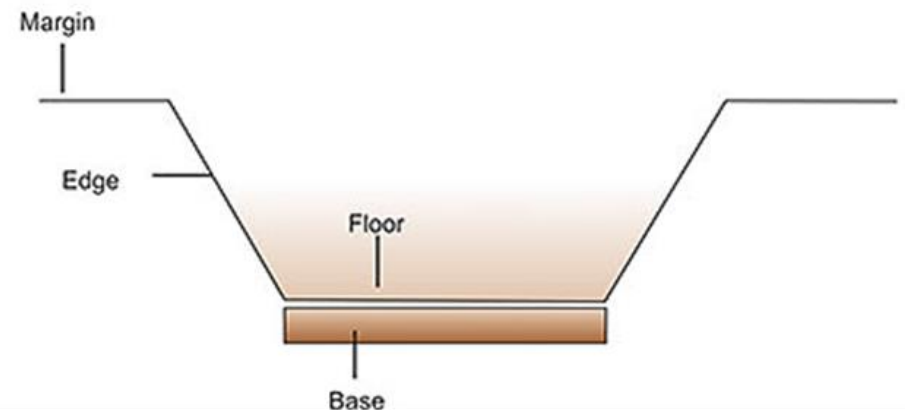
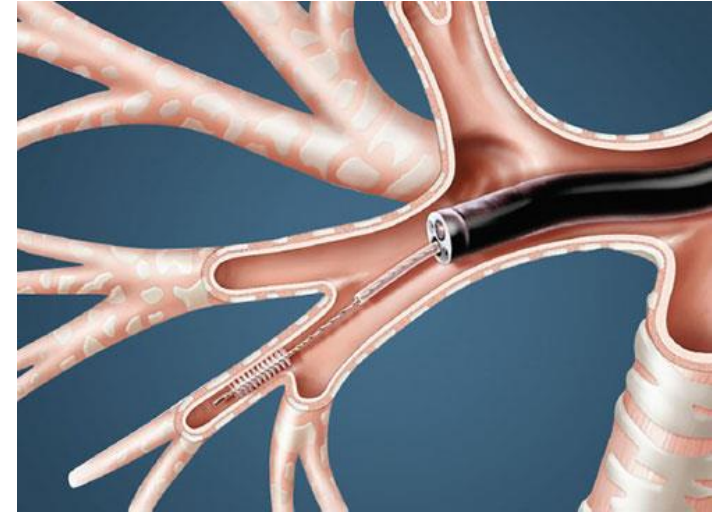
### Patient Preparation:

For gastrointestinal specimen:

To fast overnight or for at least 6 hours before the procedure.

### Procedure Concern:

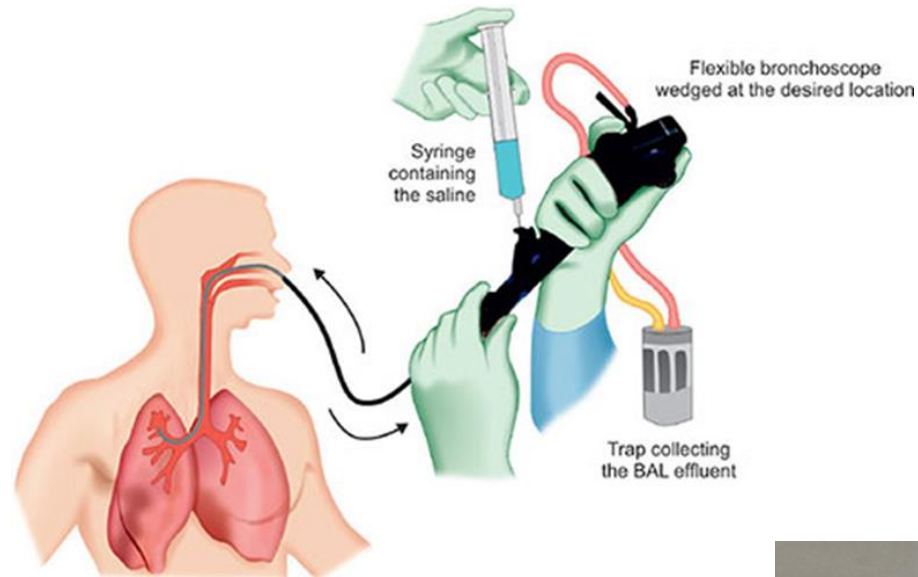
In case of ulcer, Brush the edges of an ulcer as well as the floor to obtain diagnostic material.



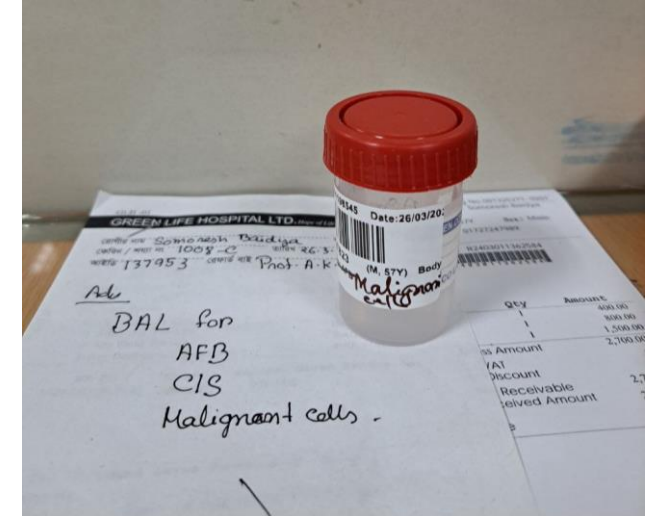
# Abrasive cyt... Washing

**Site:**

Bronchial tree



By bronchoscope – 3-5 ml balanced salt solution must be instilled before aspirating resulting material.



# ASPIRATION CYTOLOGY

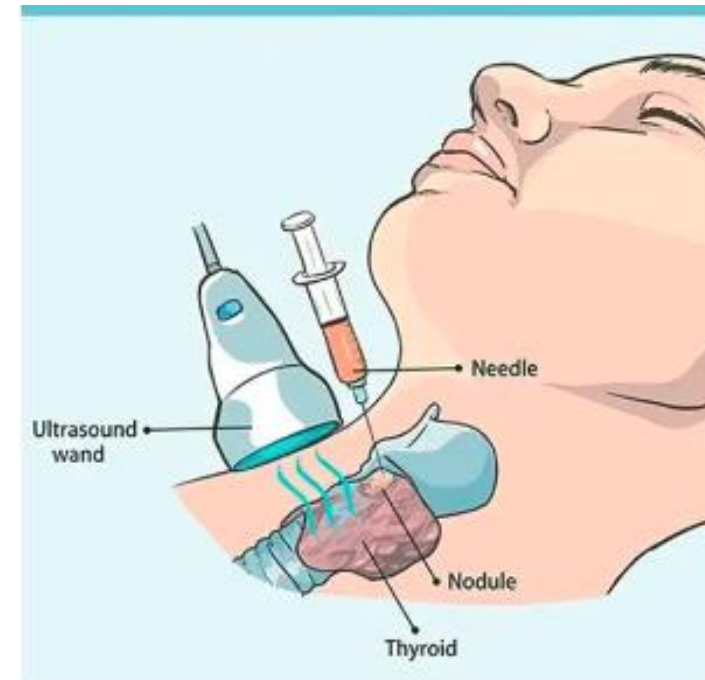
Applied in diagnosis of Palpable as well as Non palpable deeply seated lesion

## **PALPABLE MASS LESION –**

- i) Lymph node
- ii) Breast
- iii) Thyroid
- iv) Salivary gland
- v) Soft tissue masses

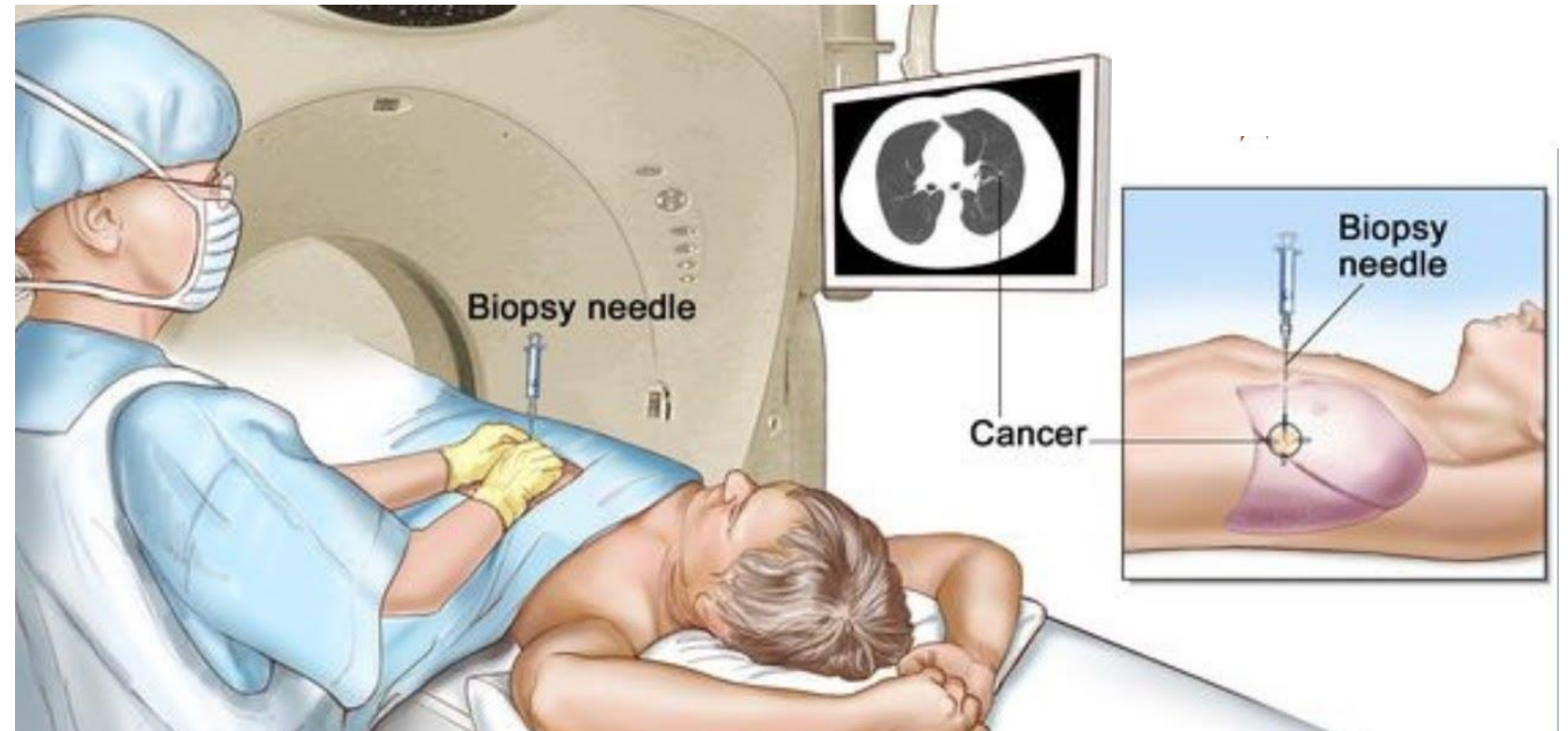
## **NON PALPABLE MASS LESION**

- i) Lung (CT guided)
- ii) Liver (USG guided)
- iii) Retroperitoneum



# Image guided FNAC procedure

- Small, non-palpable or multiple lesions.
- E.g- Thorax  
(CT guided)
- Abdomen  
(USG guided )



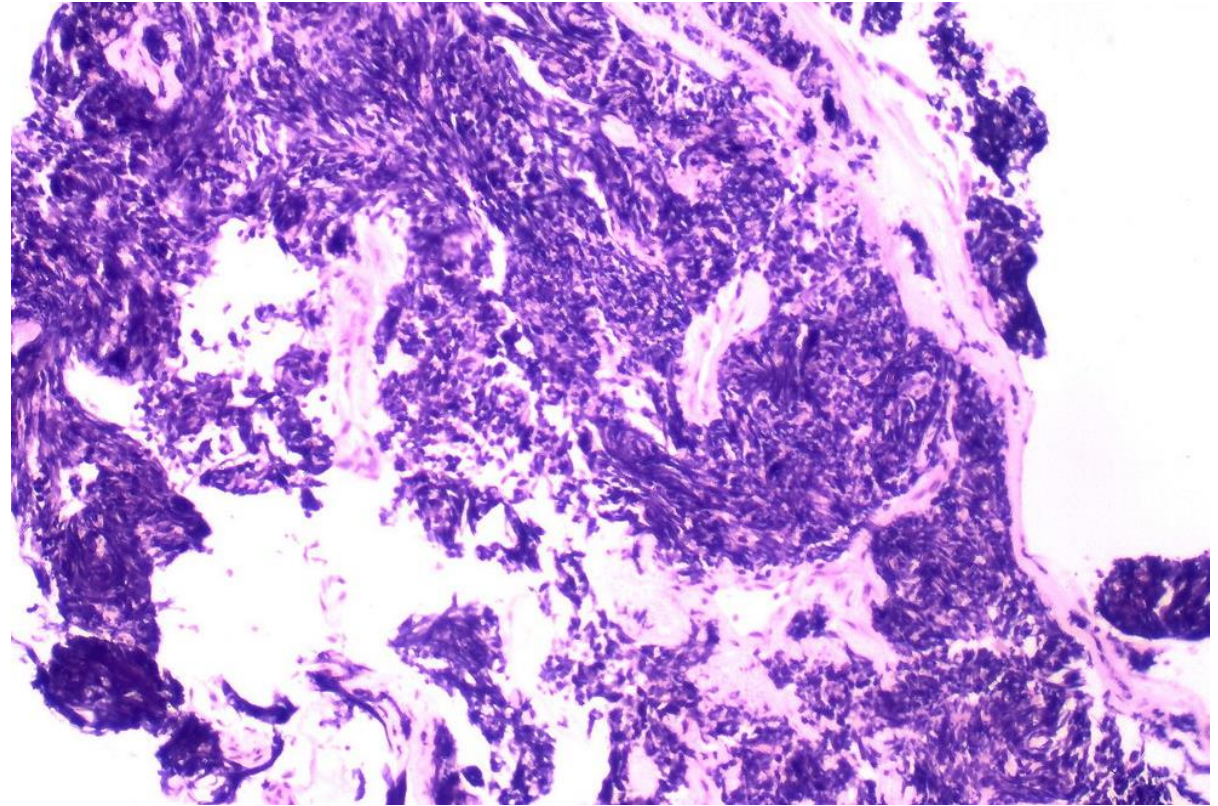
# STEPS TO AVOID ERRORS IN HISTOPATHOLOGY SAMPLE



# Step 1

## Avoid Mechanical Trauma

Gently remove the tissue to avoid trauma to the specimen caused by crushing or tearing.



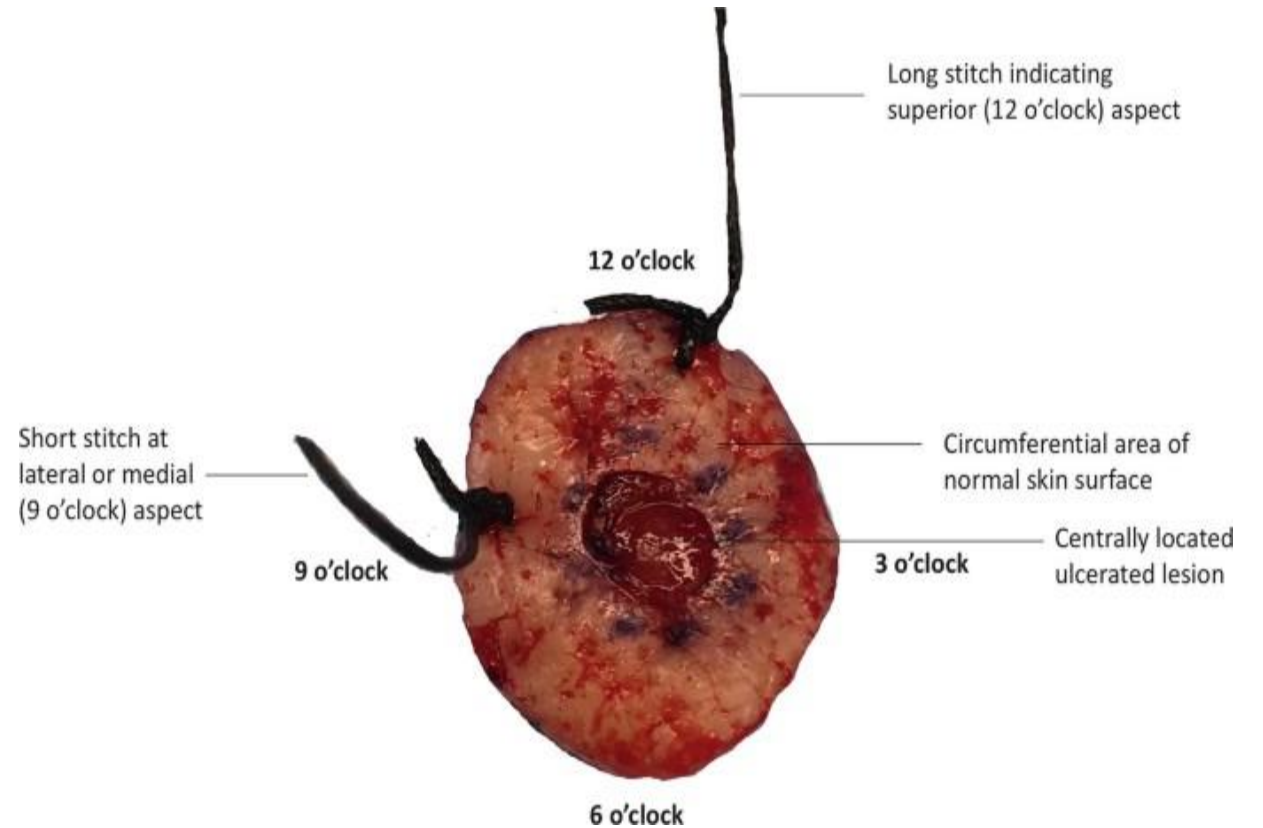
Typical crush artifact is shown in this section of lung tissue. Although no identifiable tumor cells are present, the nuclei appear dark, distorted, elongated, and intensely basophilic due to the compression caused by crushing.



# Step 2

## Denoting Surgical Margins

- Margins should be clearly excised specially in neoplasm.
- proper identification of orientation sutures must be written in request form.



# Step 3

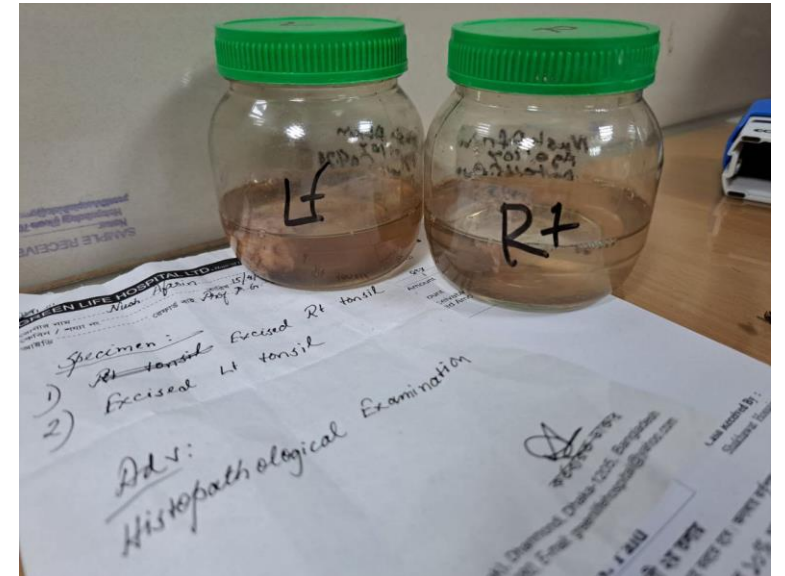
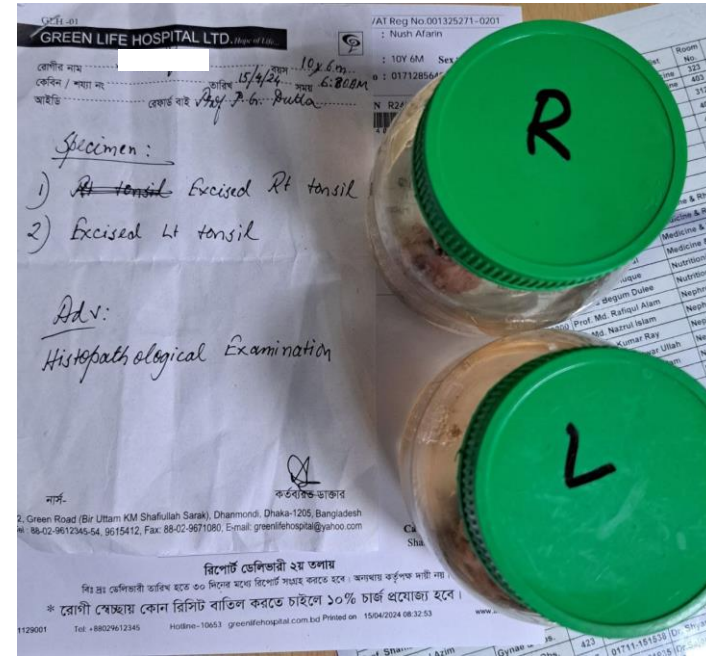
## Submitting Multiple Sites

- Submit multiple specimen of same patient in separate containers with appropriate labelling.

• Ex

Container R or Right → Tissue from right tonsil

Container L or Left → Tissue from left tonsil



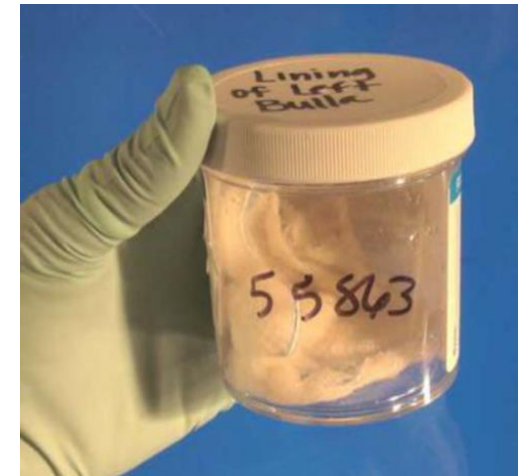
# Step 4

## Proper Handling of Endoscopic Biopsies

- Place Endoscopic biopsies on a filter paper and submerge all fragments in a same container
- Do not submit endoscopic biopsies wrapped in gauze



Using filter paper facilitates proper fixation and preserves biopsy integrity.



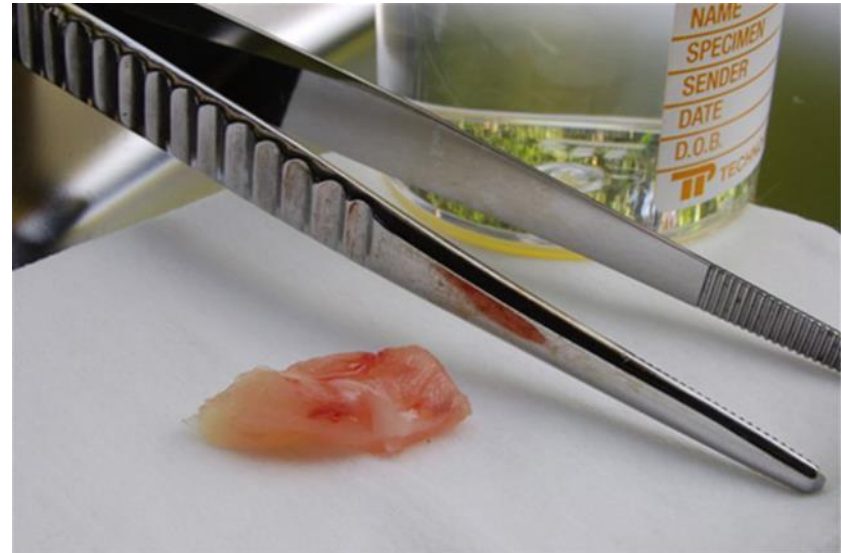
Specimen may become lost or may be crushed during attempt of retrieval process.

# Step 5

## Ensure Prompt Fixation

Process of autolysis and bacterial attack start as soon as tissue is removed from the body.

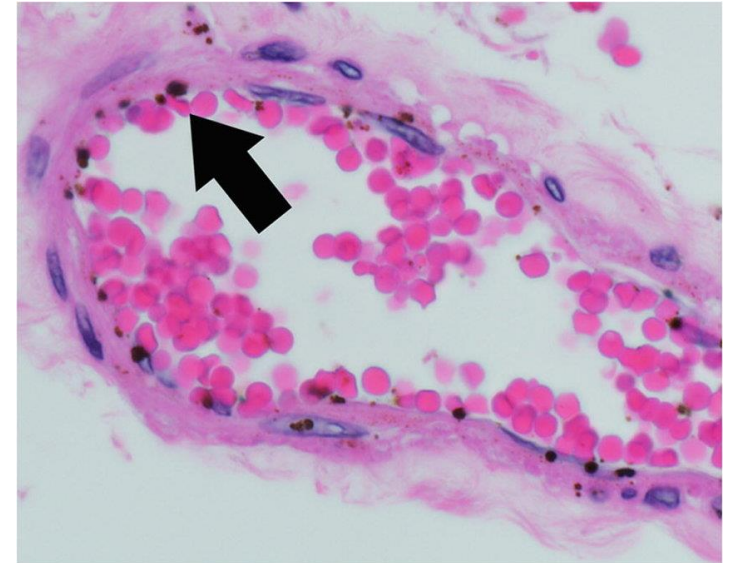
So the first aim of fixation is to arrest these changes .



To prevent degeneration or drying-out the specimen should be fixed as soon as possible.

# Ideal fixatives

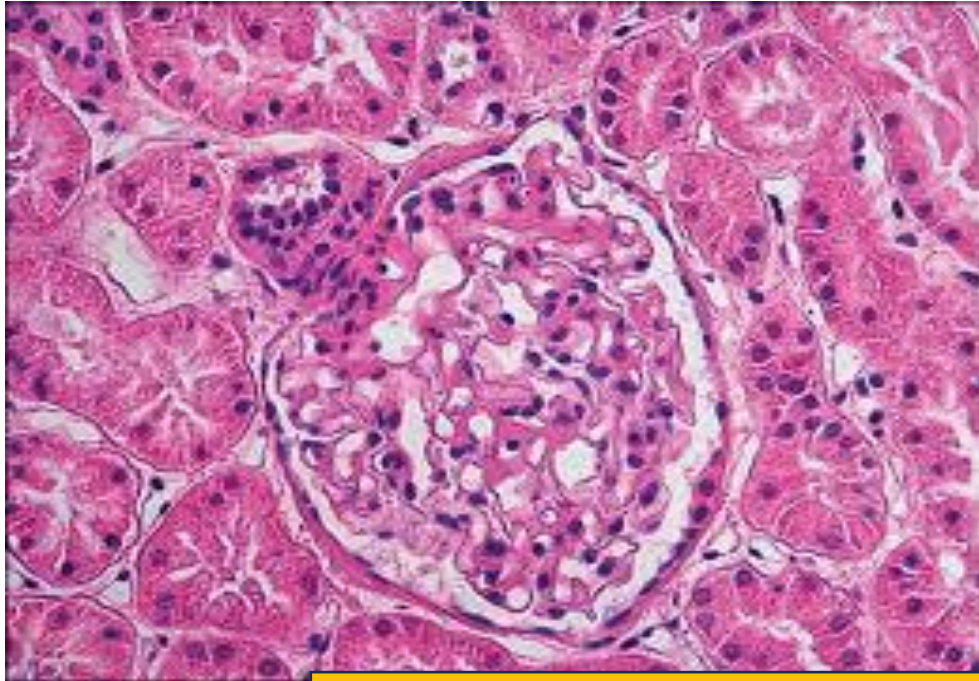
- Routine tissue fixative:  
10% Neutral buffered formalin
- High-quality fixatives with optimal pH are preferred.



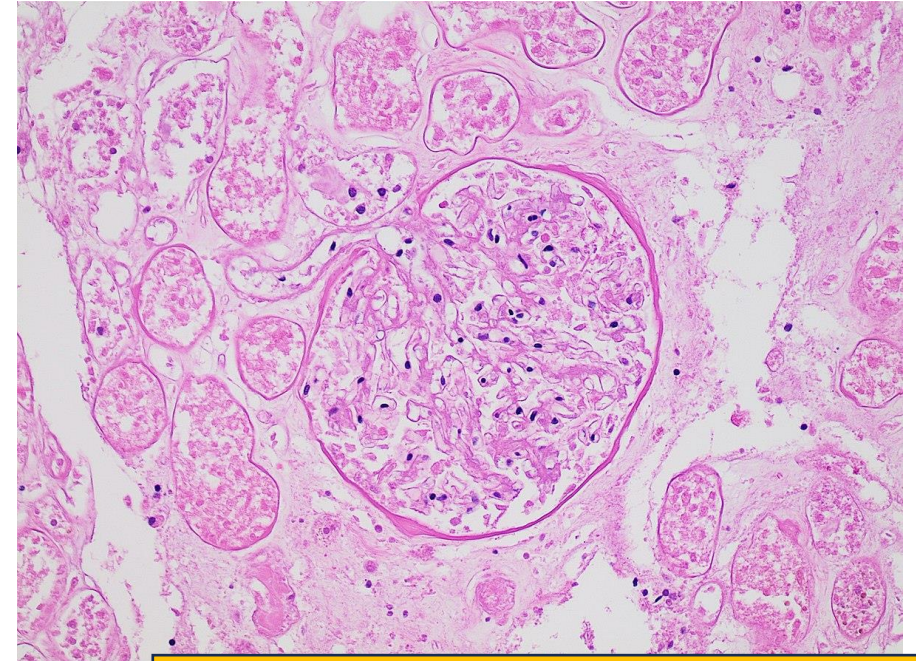
Buffered Formalin prevents formation of pigment acid  
Formaldehyde Hematin from Hb at acidic pH.

Endothelial formalin pigment  
deposition in blood vessel

- Good fixative is most important in the production of satisfactory results in histopathology



Well Preserved Renal tissue

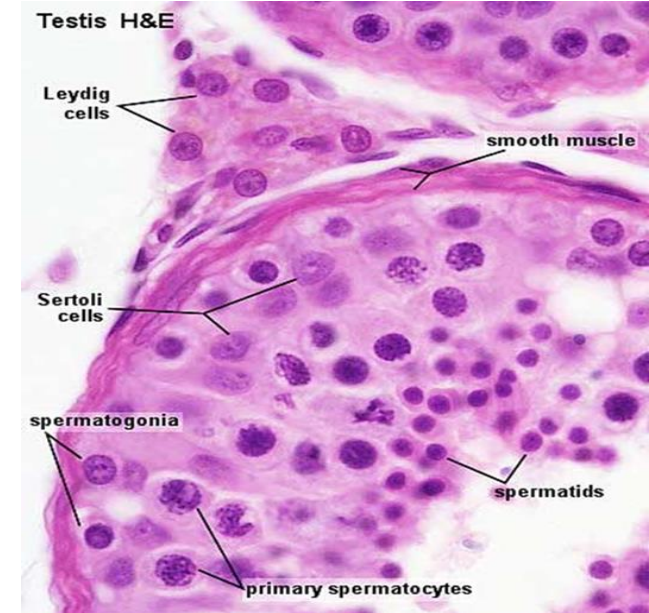


Autolysed Renal tissue

# Situations where special fixative is needed

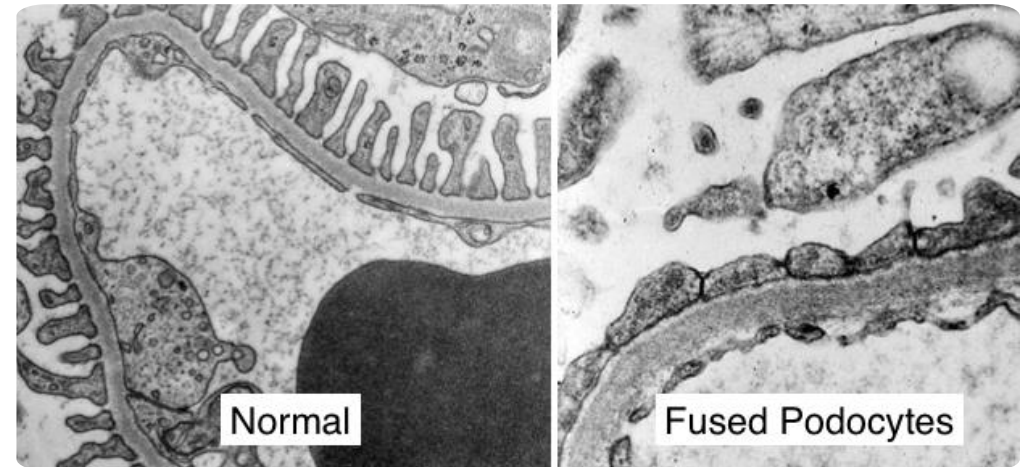
- **Testicular biopsy**

- Bouin's solution
- For better preservation of microscopic details (nucleus & chromosome) & observing cellular structures during meiosis.



- **Electron microscopy**

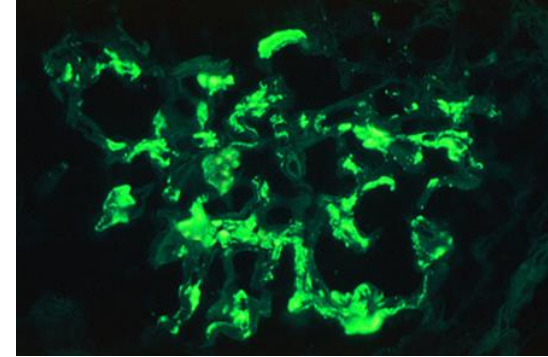
- 2 % Gluteraldehyde in 0.1 M phosphate buffer
- Example – Renal & tumor pathology



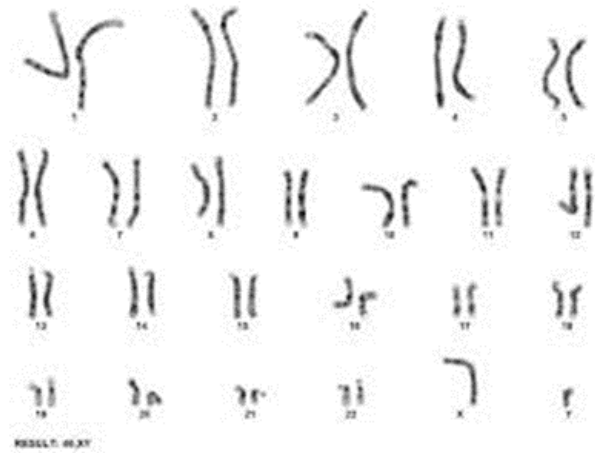
# Where fresh tissue is needed without fixatives



Frozen section



Immunofluorescence study  
– IgA nephropathy



Chromosomal study



# Frozen section biopsy

- On table diagnosis.
- Nature of lesion & surgical margin clearance.

## Pre requisition-

- Inform the pathology dept. prior to the OT day & before starting the surgery of the patient.
- Maintain the temperature on cryostat machine (-15 to -25° C)
- Fresh tiussue should be sent.



# Step-6

## Choose suitable container

- For larger specimen the container should be larger than the specimen.
- For adequate formalin penetration, ensure all biopsy specimens are fixed in a 10:1 ratio of formalin to tissue.



## Choose suitable container

- Avoid squeezing specimens into small containers.
- A small container restricts the amount of fixative, potentially resulting in incomplete fixation.



Inadequate space can lead to distortion , affecting its overall appearance and diagnostic value.



## Step-7

### Handling Large Specimens in Fixative

Large specimens that float on the fixative should be covered by a thick layer of gauze.

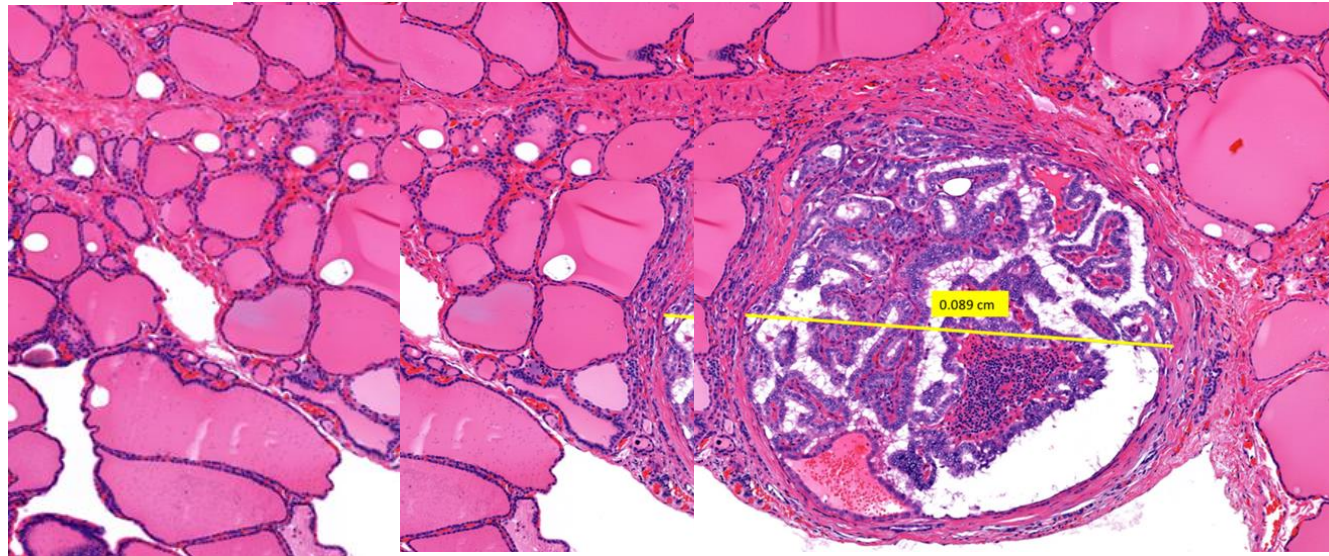


## Step-8

Submit whole specimen in a single laboratory

Don't divide specimen to submit in different laboratories.

It can make confusion to clinician as well as patient



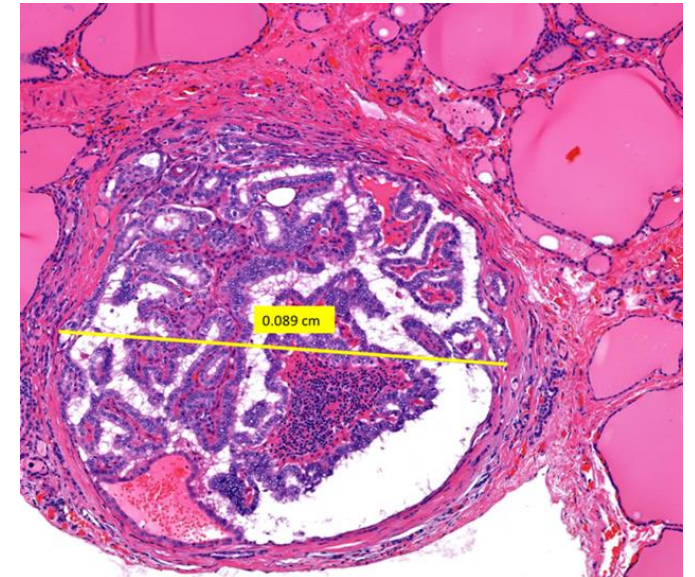
Papillary thyroid ca with MNG

A- Multinodular goiter

B - Papillary thyroid carcinoma

## Step-12..

- There can sometimes be confusion in the diagnosis when Micropapillary thyroid carcinoma is found in the background of multinodular goiter.
- This confusion arises if a surgeon divides the surgical specimen for examination by different pathologists.
- One pathologist might report Multinodular goiter, while another reports Papillary thyroid carcinoma.
- To avoid this confusion, we suggest submitting the entire specimen to one lab for a comprehensive analysis.
- If a review is needed, options are available to obtain slides, blocks, or specimens without dividing the original sample.



Papillary thyroid carcinoma in MNG

# Step 9

## Label Specimens Properly

Each specimen should be properly identified

The container should be accurately labelled including -

- Patient's full name, Age, Sex
- Word no, Bed no
- Site & side
- If more than specimen mark them as A, B, C, D etc.
- Signature of doctor with date



**Incorrect patient or specimen identification and labelling errors may lead to issuing of erroneous reports.**

Paperwork should be placed in a separate plastic bag to avoid contact with formalin if leaking does occur.





## Step 10

Avoid any unnecessary delay to send specimen to lab.



No unnecessary delays – Specimen should reach in lab within minimum time.

# Conclusion

- In this presentation, we've explored strategies for optimal patient preparation and sample collection.
- Clear communication, attention to details, and adherence to protocols reduce errors, improving reliability.
- Let's maintain high standards for better patient care.

# Sources

- Koss' diagnostic cytology and its histopathologic bases.
- Manual for Cytology
- CYTOLOGY SPECIMEN COLLECTION
- 101 steps to better histology - a practical guide to good histology practice

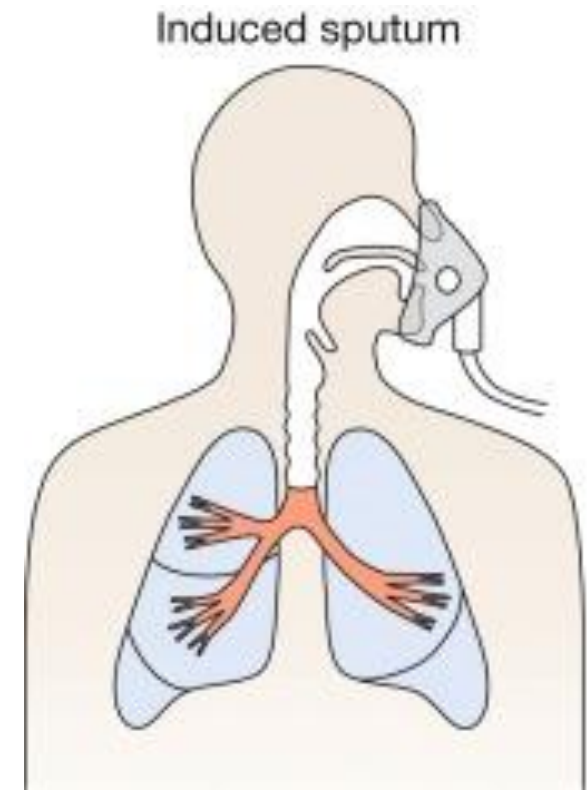


*Thank  
you!*

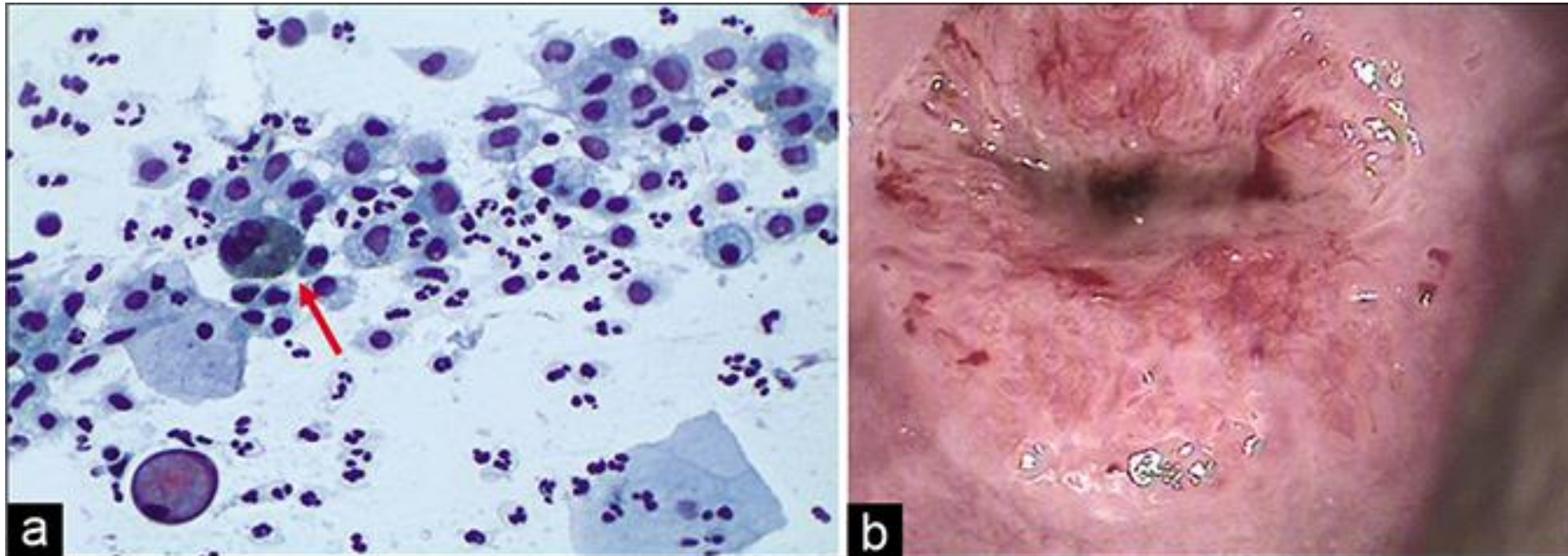


## Induced sputum

The patient inhales nebulized hypertonic saline solution, which liquefies airway secretions, promotes coughing and allows expectoration of respiratory secretions.



Abrasive cyt...  
Paps sm..



- a. CP smear – sheets of histiocyte in inflammatory background.
- b. Colposcopy showing hypertrophic edematous and congested glandular epithelium in acute cervicitis

# Abrasive cyt...

## Paps sm..

### Collection

- DO NOT use lubricant on the speculum.
- Do not clean the cervix with saline prior to collecting the sample.

✓ Causes cellular distortion or reduced number of cells

- If excessive mucus is present, then removed it by placing a small piece of sterile gauze over the cervix and gently removing it after it absorbs the exudate.



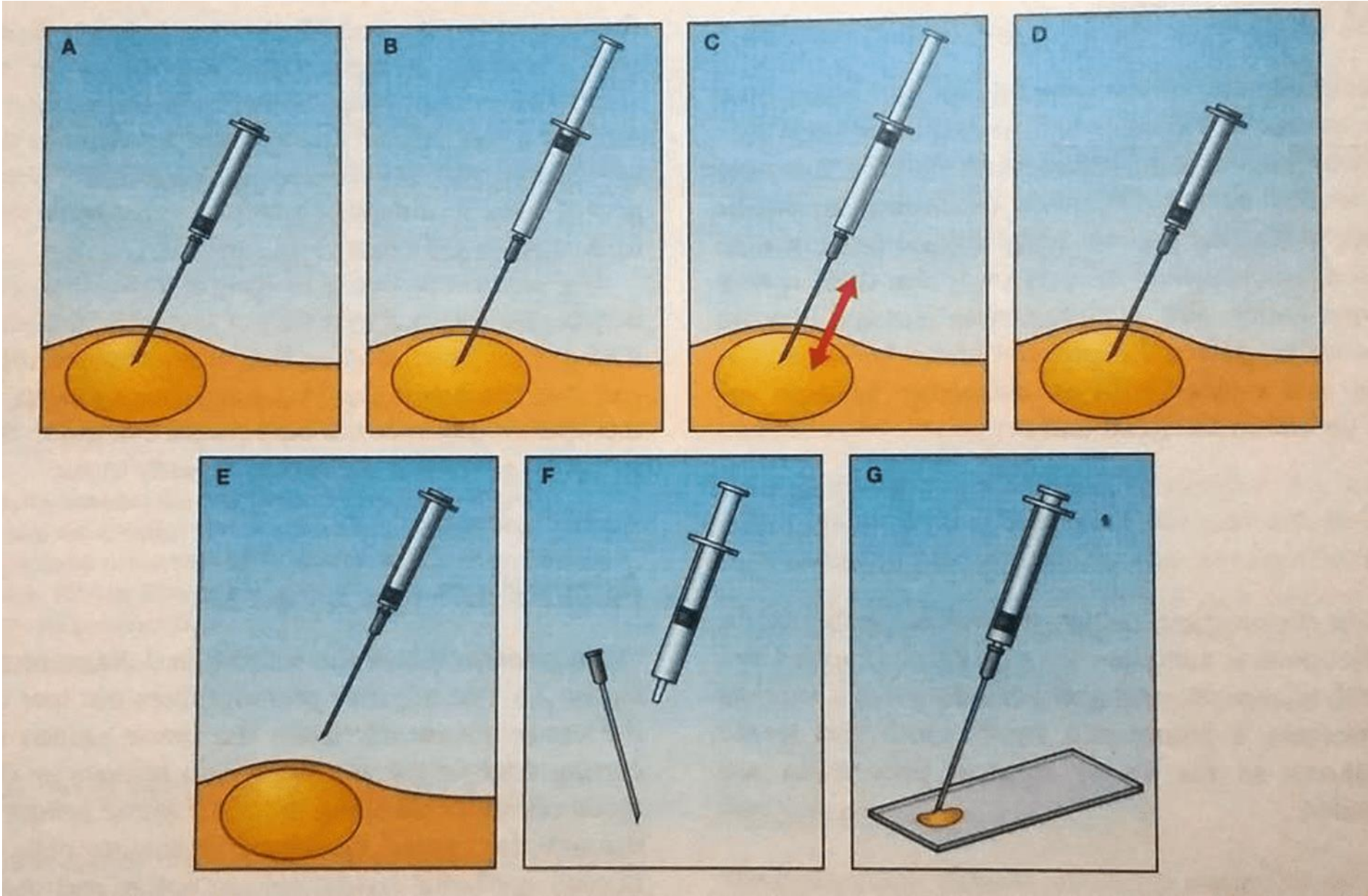
# Abrasive cyt...(back up slide)

## Skin Scrapping

- Immerse the labeled slide in a container with 95% ethyl alcohol.
- Scrape the abnormal area gently. If it's a vesicle, remove the cover and scrape both the base and the rim.
- Remove a slide from the fixative.
- Smear the collected material on the slide quickly and evenly.
- Re-immerses the slide in the fixative immediately.



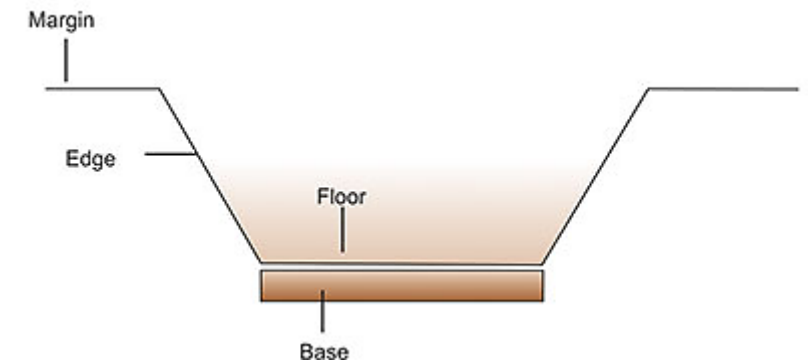
# FNAC



# Abrasive cyt... Brush..

## Procedure Steps:

- Endoscopically or Bronchoscopically directed brushing of the identified lesion.
- Brush the edges of an ulcer as well as the floor to obtain diagnostic material.
- Upon withdrawing the brush, agitate it vigorously in a 5–10 mL vial of saline or cytologic fixative.
- Do not apply the brush directly to slides.



# FNAC - benefits & limitations

## Benefits-

- Cost effective
- Outdoor procedure
- No anesthesia required
- Least invasive, minimal tissue injury
- Rapid & satisfactory diagnostic efficacy

## Limitations-

- Invasion can not be seen
- Invasive and in situ carcinoma can not be differentiated
- Skilled hand is required.

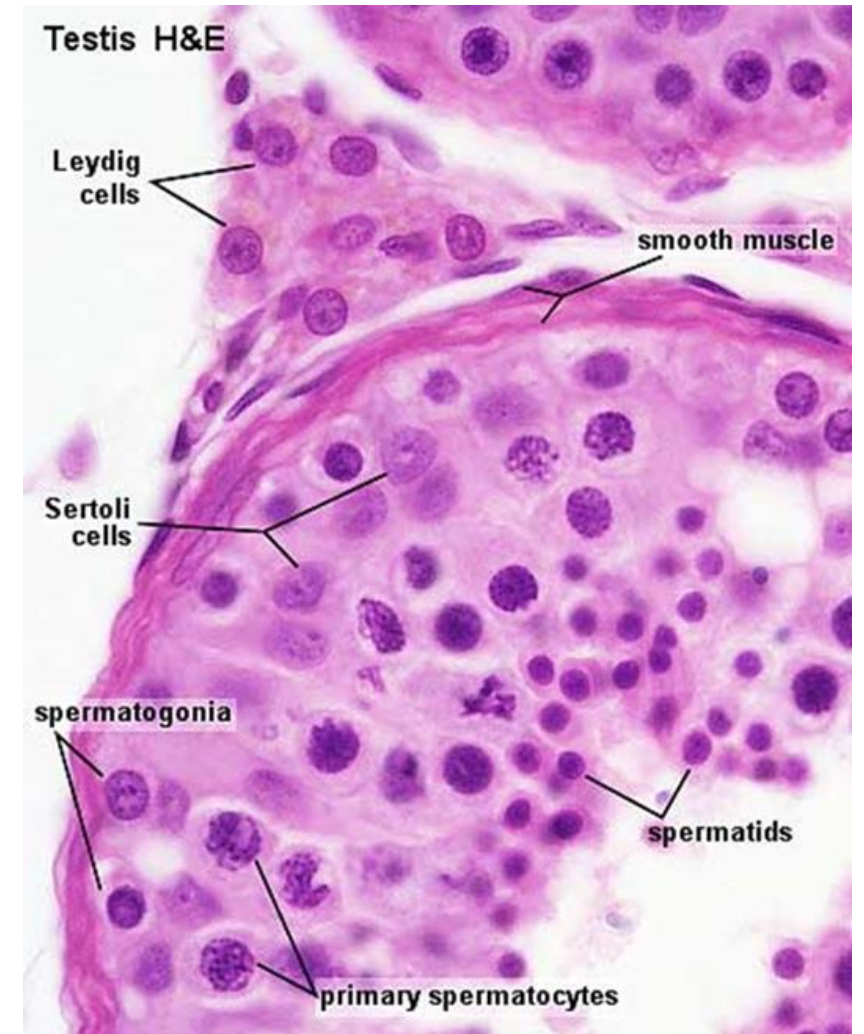
# Situations where special fixative is needed

- Testicular biopsy

Bouin's solution

For better preservation of microscopic details-

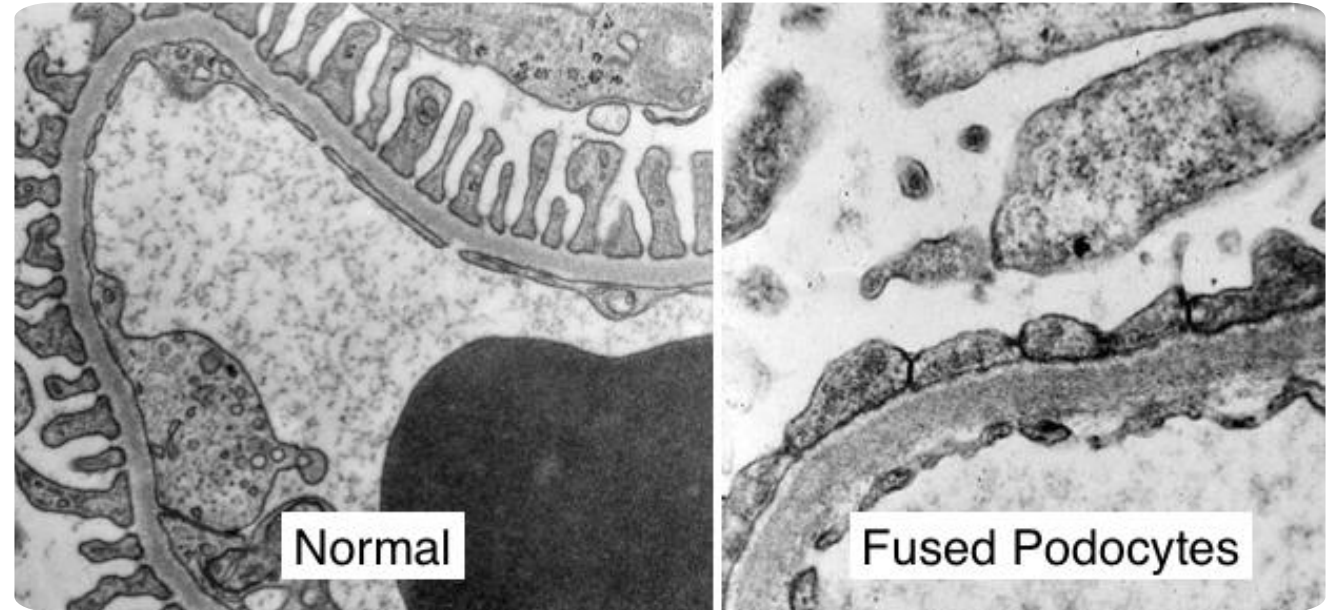
- ✓ It effectively preserves the nuclei and chromosomes within the testicular tissue.
- ✓ This is especially important when observing cellular structures during meiosis.



## Situations where.....

### Electron microscopy-

- 2 % Gluteraldehyde in 0.1 M phosphate buffer
- Example – Renal & tumor pathology



# Frozen section.



## Pre requisition-

- **Advance Notification:**

Notify the pathology department in advance, both before the patient's surgery day and prior to the start of the surgery.

This ensures that the necessary arrangements are made for the frozen section procedure.

- **Cryostat Temperature:**

Maintain the cryostat machine at a temperature range of  $-15^{\circ}\text{C}$  to  $-25^{\circ}\text{C}$

The cryostat is used for cutting frozen tissue sections, and maintaining the correct temperature is crucial for optimal results

# Frozen section.



- **Fresh Tissue:**

Ensure that fresh tissue is sent for the frozen section.

Fresh tissue provides the most accurate representation for immediate evaluation during surgery.

- **Effective Communication:**

Maintain clear communication between the surgeon, pathologist and Share relevant clinical information (e.g., patient history, radiology findings) with the pathologist.

This context helps guide the interpretation.



# TAKE HOME MESSAGE...

- ❖ It is human to make error, But it's also human to react and create solutions.
- ❖ So precise and correct actions should be taken to control pre-analytic and post-analytic errors.
- ❖ We should never be relaxed or overlook these errors.
- ❖ This is the only way for continuous quality improvement for better patient care.