

Alnoor University College
Practical Physiology lecture
Anesthesiology Techniques Department
First stage



كلية النور الجامعة
محاضرة الفلسفة العملية
قسم تقنيات التخدير
المرحلة الاولى

Microscope

(Lab:1)

Assistant lecturer: Amina J. Mohammed

Introduction:

The microscope is one of the most commonly used instruments in medical and other laboratories .

The word **microscope** comes (from the Ancient Greek: μικρός, mikrós, "small" and σκοπεῖν, skopeîn, "to look" or "see") is an instrument used to see objects that are too small to be seen by the naked eye.

Objectives:

At the end of this session the students should be able to describe:

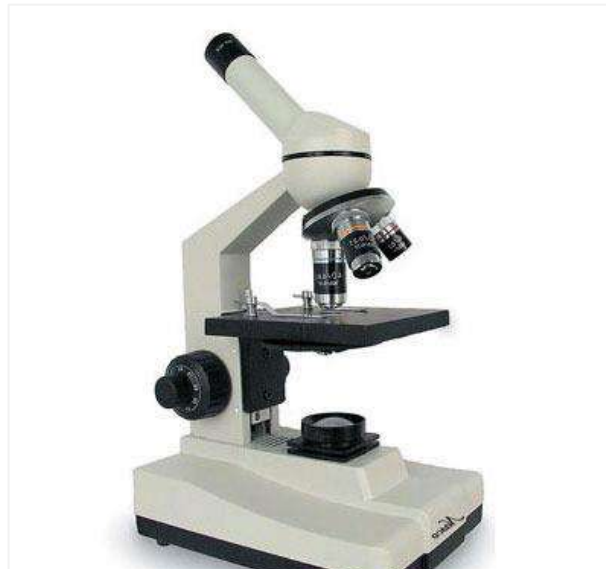
1. Structure and the parts of compound microscope.
2. How to use the microscope efficiently.

Types of Microscopes

1. Simple microscope.
2. Compound light microscope.
3. Electron microscope; this includes:
 - A. Transmission electron microscope (TEM).
 - B. Scanning electron microscope (SEM).
4. Phase-Contrast microscope.
5. Interference microscope.



Simple microscope



Compound light microscope



Transmission electron microscope



Scanning electron microscope



Phase-Contrast microscope



Interference microscope

Parts of Microscope

1- The Support System:

- A. Base:** It is a heavy metallic , u- or horseshoe-shaped base or foot , which supports the microscope on the worktable to provide maximum stability.
- B. Handle (The arm or limb):** It supports the body tube;it is used to carry the microscope.
- C. Body Tube:** fitted at the upper end of the handle, either vertically or at an angle, the body tube is the part through which light passes to the eyepiece , thus conducting the image to the eye of the observer. It is 16-17cm in length.
- D. The Stage:** it has two components: the **fixed stage** and the **mechanical stage**.

Fixed stage: it is a square platform with an aperture in its center, and fitted to the limb below the objective lenses. The slide is placed on it and centered over the aperture for viewing.

Mechanical stage: it is a calibrated metal frame fitted on the right edge of the fixed stage.

2- The Focusing system:

consists of :

A. Coarse adjustment.

B. Fine adjustment.

The **coarse adjustment** knob should only be used with the lowest power objective lens.

Once it is in focus, you will only need to use the fine focus.

Fine adjustment: Fine tunes the focus and increases the detail of the specimen. Using the coarse focus with higher lenses may result in crashing the lens into the slide.

3- The Optical (Magnifying) system:

consists of **the body tube** , **the eyepiece** , and **the nosepiece** that carries **the objectives**.

A. Body tube: Connects the eyepiece to the objective lenses.

B. Eyepiece: The lens the viewer looks through to see the specimen. The eyepiece fits into the top of the body tube. Usually contains a **10X** or **15X** power lens.

C. Nosepiece: A rotating turret that houses the objective lenses. The viewer spins the nosepiece to select different objective lenses.

D. Objective lenses: One of the most important parts of a compound microscope, as they are the lenses closest to the specimen they are low- power(4x,10x) and high power (**40x,100x or oil immersion**).

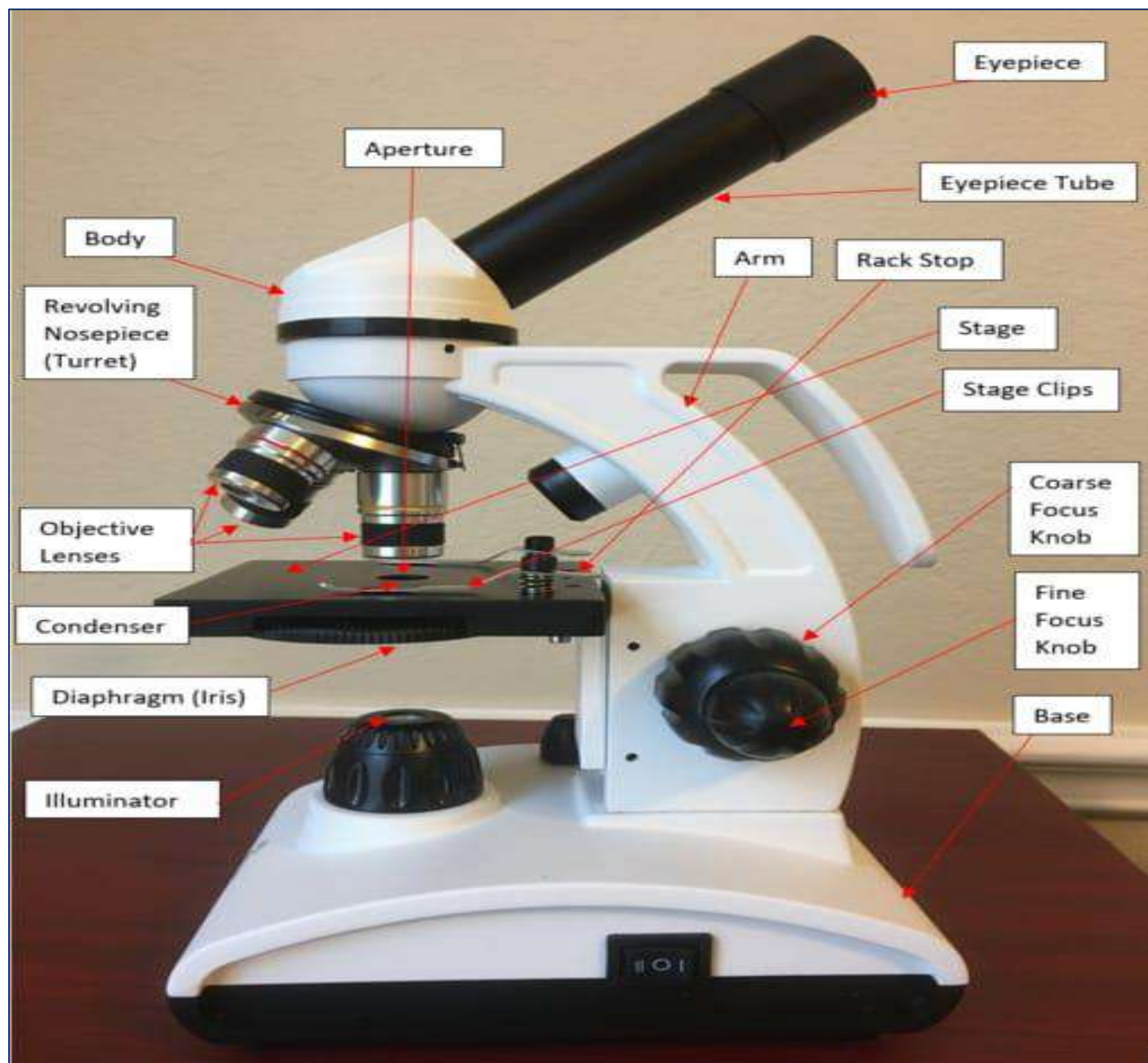
4- Illumination system:

A. Light source.

B. Condenser: gathers and focuses light from the illuminator on to the specimen being viewed.

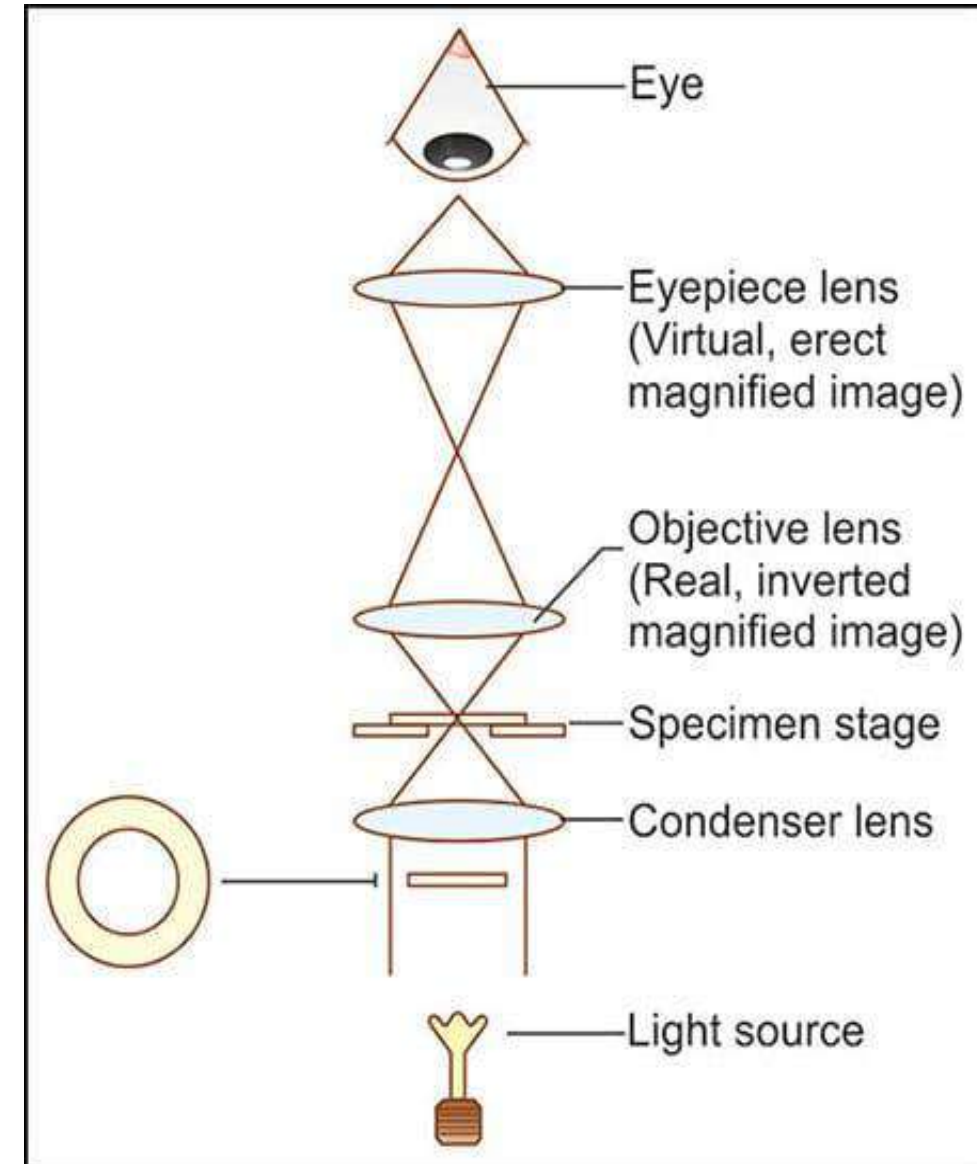
C. Iris diaphragm: Adjusts the amount of light that reaches the specimen.

D. Filter.



How Does a Microscope Work?

All of the parts of a microscope work together. The light from the illuminator passes through the aperture, through the slide, and through the objective lens, where the image of the specimen is magnified. The magnified image continues up through the body tube of the microscope to the eyepiece, which further magnifies the image the viewer then sees.



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Blood samples & their collection methods

(Lab:2)

Assistant lecturer: Amina J. Mohammed

Blood samples:

The term “blood sample” refers to the small amount of blood (a few drops or a few milliliters) obtained from a person for the purpose of testing or investigations. These tests are carried out for aiding in the diagnosis of the disease or disorder.

Types of blood samples:

- ❖ Whole blood.
- ❖ Serum.
- ❖ Plasma.

Objectives:

At the end of this session the students should acquire:

- The skills of collecting blood samples, **Venous** and **Capillary**.
- Knowledge how to blood separate of cell, plasma and serum.

Collection of blood Samples

There are **three** essential methods for the collection of blood sample's:

1- Vein Blood Sampling:

When a large sample is required, this can be obtained by venipuncture and placed in a container that contains an anti-coagulant substance such as EDTA (Ethylene Diamine Tetra Acetic acid). This method is the most common of collecting blood from adults.

Note: We need to collect blood from a superficial vein in the upper limb; **the median cubital vein** is the most used to do so, since it is close to the skin and does not have many nerves nearby.

Venous blood is necessary is used for certain tests such as: **Estimation of ESR, Estimation blood constituents like urea etc.. , bacteriological and serological examination.**

2- Arterial blood sampling:

The **radial artery** is used for blood collection. This artery is located on the thumb side of the wrist. Since it is small, the radial artery needs a professional person to collect the blood of the patient.

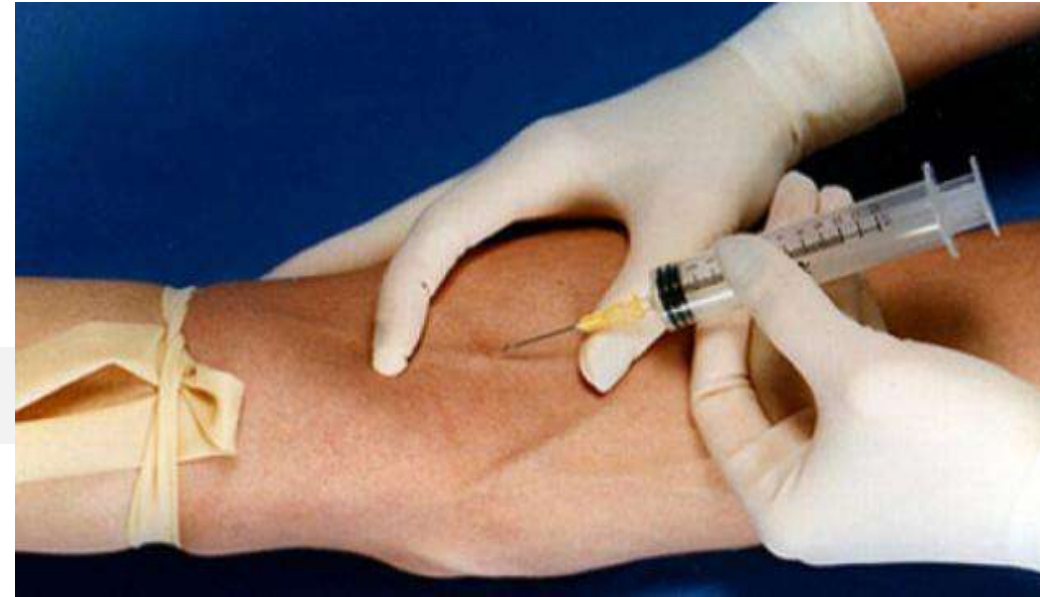
We can also depend on other arteries in the blood collection like the **brachial** and the **femoral artery**, but these arteries have certain **disadvantages** such as:

- ❖ Being difficult to find, because they are less superficial than the radial artery.
- ❖ They are surrounded by structures that could be damaged by faulty technique.

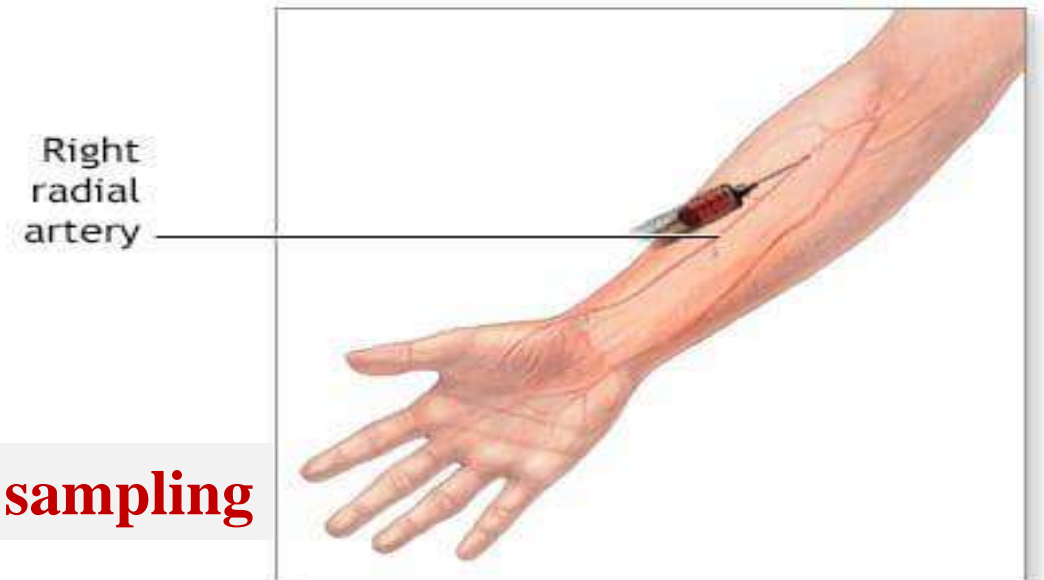
Arterial blood is necessary is used for certain tests such as: **blood pH, gas levels, etc..**



Vein Blood Sampling



Arterial blood sampling



3- Capillary blood sampling:

Small samples can be obtained from blood by pricking a **finger-tip** or **ear-lobe** or **heel**. may be performed on patients of any age, they are used for specific tests that need small quantities of blood, such as:

1. Estimation of Hb, RBC, WBC and Platelet counts.
2. Preparation of peripheral smear.
3. Blood grouping
4. Estimation of blood glucose.

Procedure

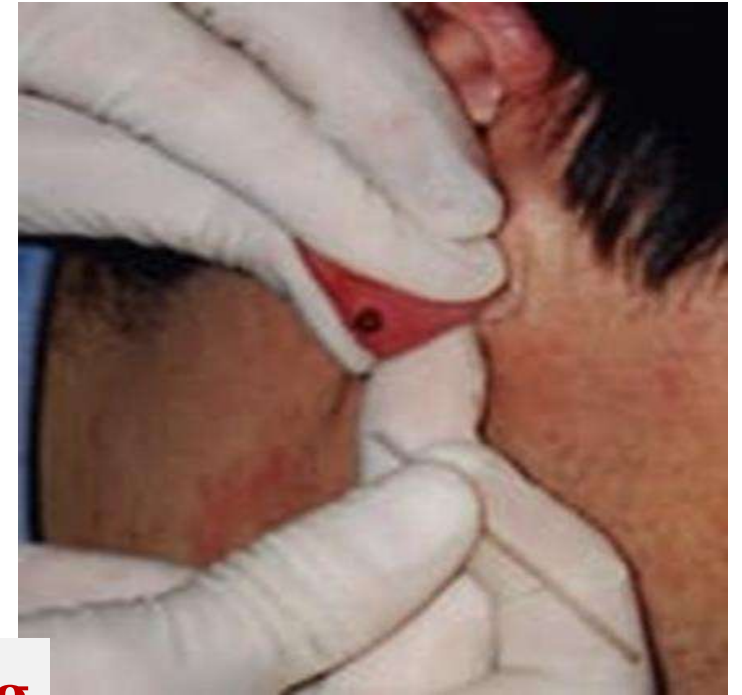
- ❖ The finger to be punctured must be free of any visible infection or wound; it should be massaged to increase circulation before being punctured with a sterile disposable lancet.
- ❖ Wipe the finger-tip or ear-lobe with an alcohol-damped swab and allow to dry in air (never blow). If the finger is not dry, the blood will spread and be difficult to sample.
- ❖ Puncture deeply (3mm deep) with a sterile disposable lancet, a free flow of blood is essential.
- ❖ Use a new lancet for each puncture.

Note: Puncturing the fingers of infants younger than 1 year of age is not recommended. Puncturing of the heel or toe may be more suitable for infants.

Filling Capillary Tubes:

Usually, capillary tubes are not provided with automatic filler. Therefore, all you need is to hold the tube between fingers and apply it to the flowing blood at 45° the blood flows into the capillary tube spontaneously according to the capillary phenomenon, no need to aspirate. Applying the tube horizontally might trap air bubbles into the sample.





Capillary blood sampling



Pipetting Blood:

A blood pipette is a straight capillary pipette with a single graduation mark at 20 or 50 microliters (0.02 or 0.05 ml), which is attached to a rubber pipette.

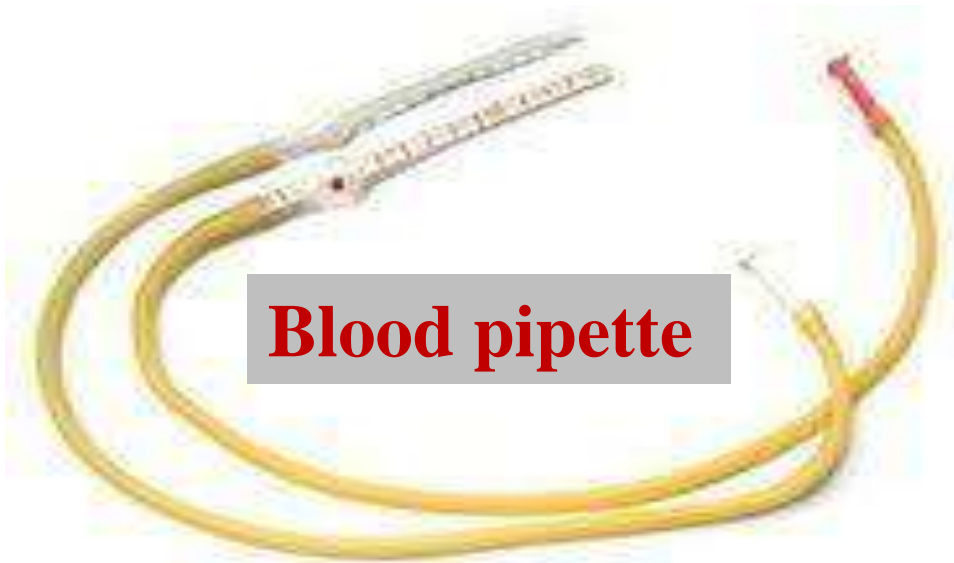
Expel the blood into the appropriate reagent solution and rinse the pipette two to three times with the solution to wash any blood left in the pipette into the suspension. you might need to do dilutions so you need a larger pipette.



Plastic pipette



Hb pipette



Blood pipette



Blood separation to cell, plasma and serum

Blood: is a complex fluid consisting of plasma—extracellular fluid rich in proteins and of formed elements—red blood cells (RBCs), white blood cells (WBCs), and platelets. Total blood volume is **70** mL/kg body weight in the adult woman and **80** mL/kg body weight in the adult man.

Plasma: is a pale-yellow solution of electrolytes, plasma proteins, carbohydrates, and lipids. Pink-collared plasma suggests the presence of haemoglobin caused by haemolysis (lysis of RBCs) and release of haemoglobin into the plasma. The normal plasma volume is 55% of body weight or of total blood volume.

Serum is the fluid that remains after whole blood is allowed to clot and the clot is removed. The serum has essentially the same composition as plasma except that:

1. Clotting factors: I-fibrinogen, II-prothrombin.
2. It has higher serotonin content because of the breakdown of platelets during the clotting mechanism.

Preparation of plasma and serum

Separation of plasma:

Blood will be collected into purple top **EDTA tubes** and centrifuged (2000-3000 rpm) for 15 minutes.

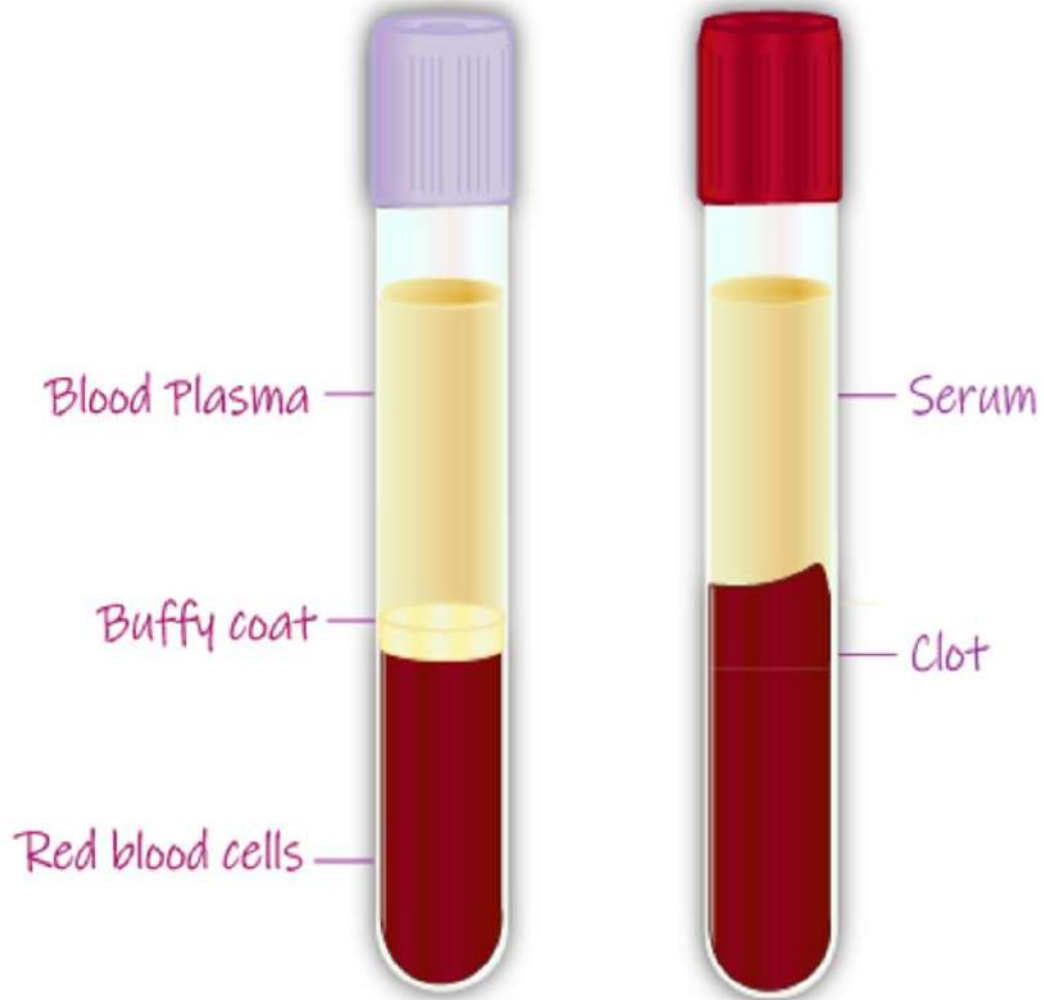
Separation of serum:

whole blood will be collected following standard procedures using a serum separator tube, Allow samples to **clot for** 15-30 minute at room temperature, centrifuge for 15 minutes at approximately 2000-3000 rpm.

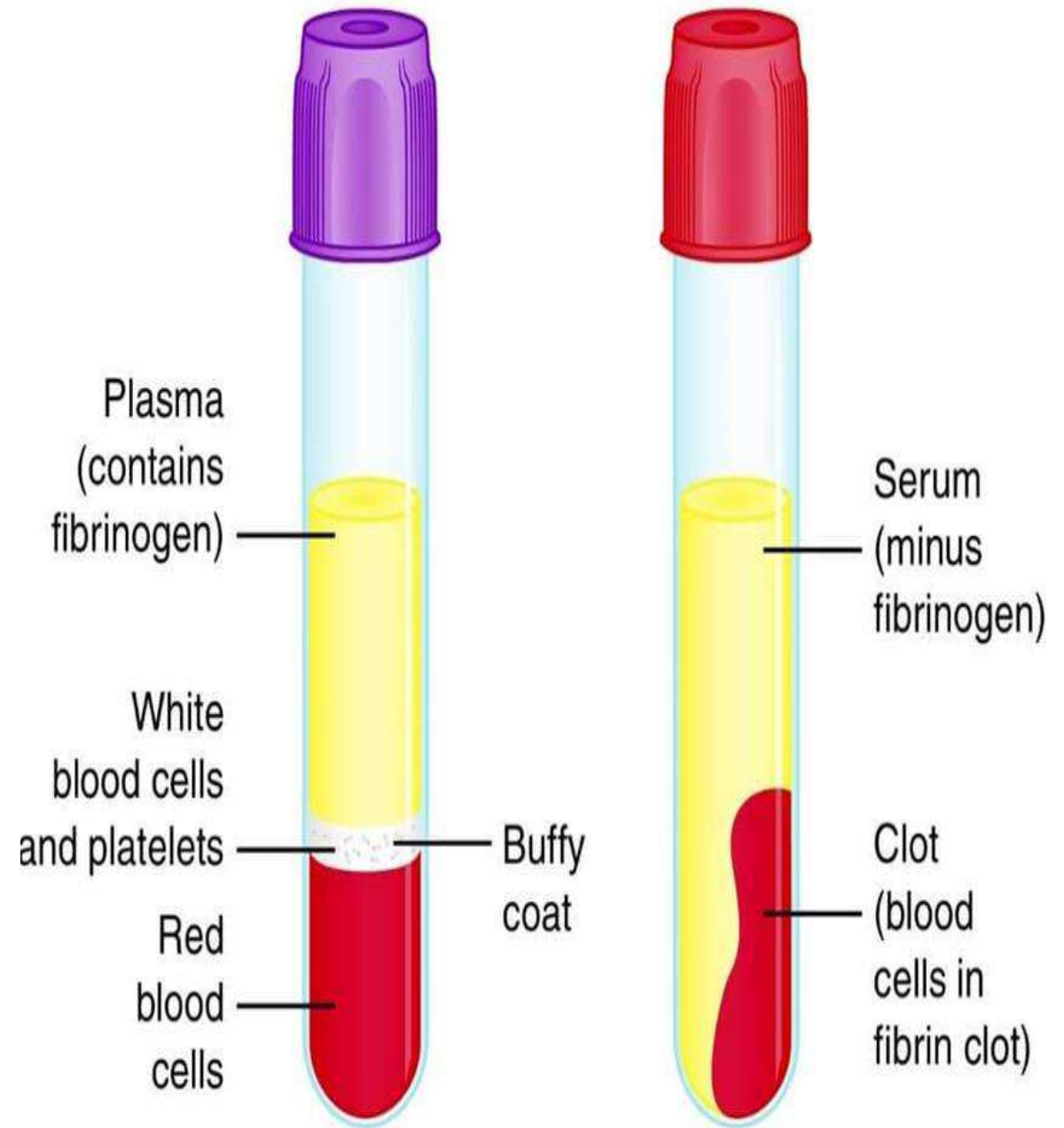
Centrifuge



Plasma vs Serum



Serum = Plasma - fibrinogen



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Hemoglobin Estimation

(Lab: 3)

Assistant lecturer: Amina J. M. Al-hayani

Introduction and Principle

Hemoglobin is the pigment inside the red blood corpuscles which carries oxygen from the lungs to the tissues. the amount of hemoglobin can be estimated by different methods, mostly based on the intensity of colour or light absorption of a stable derivative of hemoglobin.

Hb is a protein in nature composed of two portions:

- **Heme:** the heme consists of iron and porphyrin ring.
- **Globin:** which consists of two pairs of chains α and β .

Types of Hb:

- 1.Hb A:** consists of 2 α and 2 β .
- 2.Hb F** (F present in infants): consists of 2 α and 2 γ .

Objectives:

1. At the end of this session, the students will be able to estimate hemoglobin concentration in a provided blood sample.
2. Differentiate between males and females regarding hemoglobin concentration.
3. Attendees gain the knowledge and skills necessary to develop their own capabilities in assessing disease conditions.

Materials and Methods

1. Electric cell counter.
2. Manual method.

A- Sahli's method:

Is the most primitive unreliable method simply because of the human error and personal feeling. The results therefore might divert some 10-20% from the true results that can never be accepted.

Principle:

1. To convert of (Hb) into acid haematin.
2. Dilute the acid hematin by distilled water.
3. Compare hematin color with standard color in comparator block.

Devices and Solution used:

- ❖ Hemoglobinometer.
- ❖ Capillary blood (tip of the finger).
- ❖ HCL 0.1N.
- ❖ Distilled water.
- ❖ Dropper, glass rod (Stirrer).

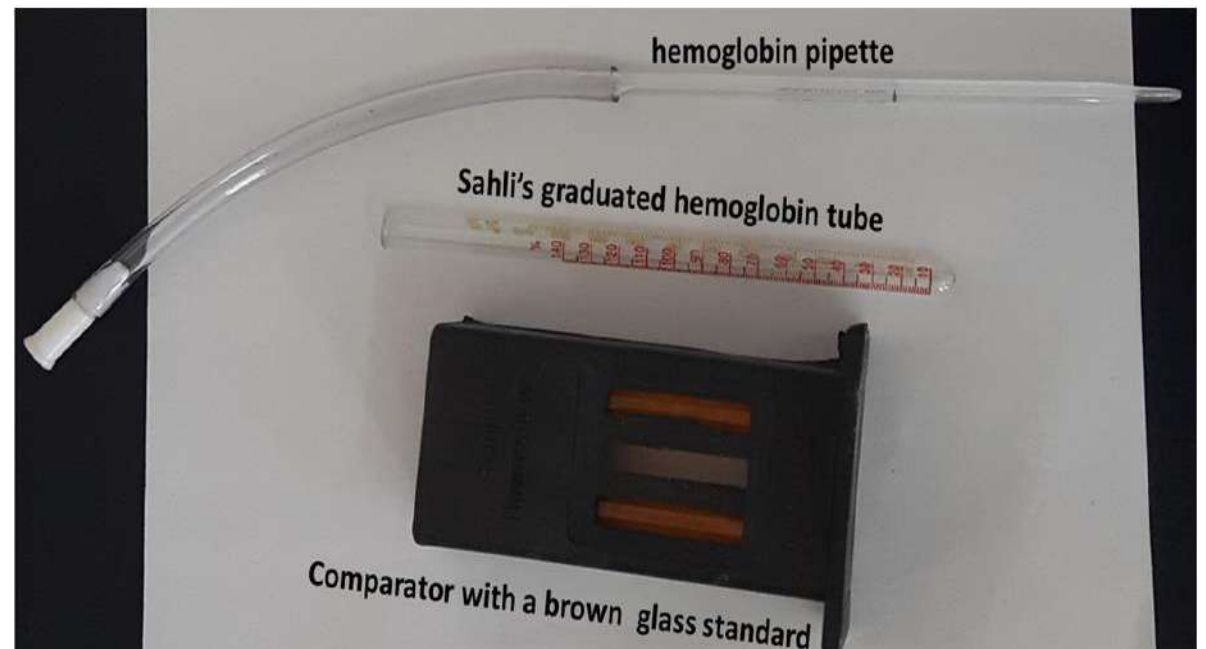


Figure (1): Haemoglobinometer (sahli method).

Procedure:

1. Place (0.1N) HCL to the 10% mark of (5 drops) of graduated sahil tube using pipette, because it hydrolysis all types of Hb to form acid haematin.
2. Clean the tip of finger and then the blood is sucked by Hb pipette up to 20 mm³.
3. Mix the blood and HCL by stirring with a glass rod, and then let the tube stand for 10 minutes.
4. Place the tube in the comparator block and hold it up to strong light. Add distilled water drop by drop to the hemation solution (stir) until its color matches the color of the standard color on the comparator.
5. Read yellow the scale on the sahli tube to obtain the percent of Hb per 100 ml of blood.

Note:

HCL used to (Benefits of HCL):

A- Prevent the coagulation of blood.

B- Brake up the cell wall, and Hb release.

When mixed with Hb to yield acid hematin, a brownish colored solution



☐ Don't press the finger during blood drawing.

B- Colorimetric method:

Is a more accurate and reliable method.

Principle:

When cyanmethemoglobin reagent (drabkin's solution) containing **potassium cyanide** and **potassium ferricyanide** is added to the provided blood sample the **ferricyanide** will convert hemoglobin iron from the ferrous (Fe^{2+}) to the ferric (Fe^{3+}) state to form **methemoglobin**. the latter then combines with potassium cyanide to give the stable pigment **cyanmethemoglobin**.

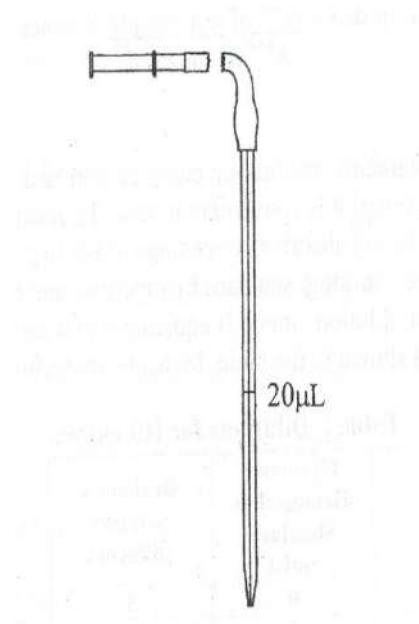
The absorbance of this pigment is measured with a spectrophotometer at a wavelength of **540 nm**.

Devices and reagents:

- Spectrophoto-meter.
- Drabkin's solution.
- Sterile lancets.
- Hb pipette



**Figure (2): Spectrophotometer
(Colorimetric method)**



Hb pipette 20µL.

Procedure:

1. Add 2.5 ml of the reagent (Drabkin's solution) into both the blank and (test) tubes.
2. Add 10 μ l of blood into the (test) tube.
3. Rinse the pipette several time.
4. Mix well and allow to stand for 5 minutes.
5. Use the blank tube to adjust the colorimeter to zero optical density at 540 nm wave length.
6. Read the optical density of the test.

Results and Calculations:

The concentration of hemoglobin in g/dl can be estimated either by: Beer's law

$$\text{Concentration of Hb (g/dl)} = \frac{A \text{ of test sample}}{A \text{ of standard}} \times \text{concentration of standard}$$

A = absorbance

Or:

The concentration of hemoglobin in g/dl can be estimated Directly read from standard calibration curve or standard table.

Table (1): Dilutions for Hb curve.

Tube Labeling	Cyanmet-Hemoglobin Standard (mL)	Drabkin's Solution (diluent)	Hemoglobin Concentration (g/dL)
Blank	0	5	0
1	1	4	4
1 duplicate			
2	2	3	8
2 duplicate			
3	3	2	12
3 duplicate			
4	4	1	16
4 duplicate			
5	5	0	20
5 duplicate			

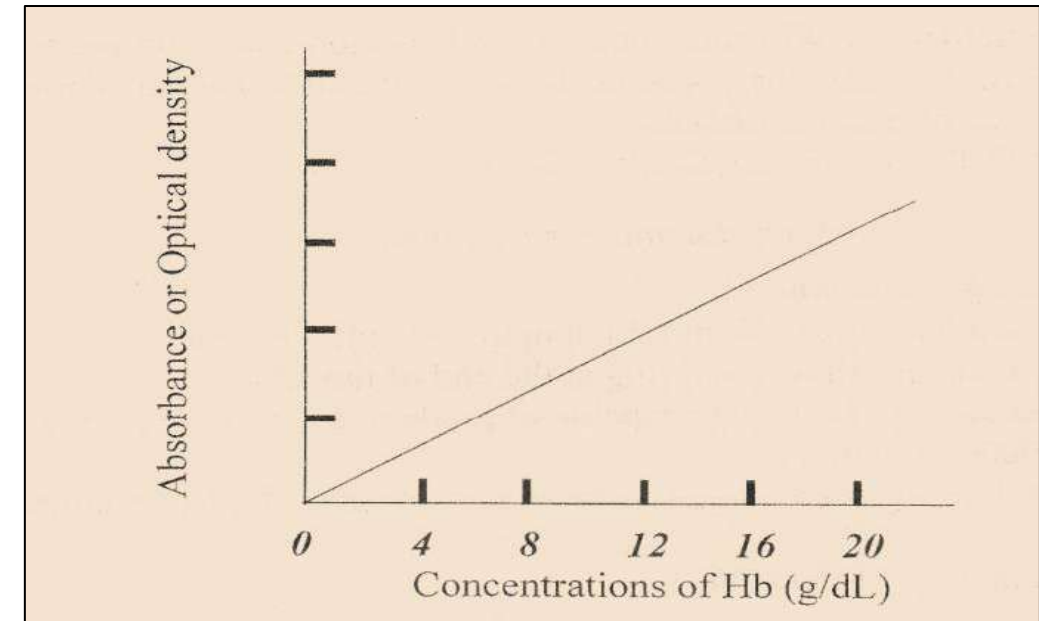


Figure (3): Hemoglobin calibration curve.

Normal hemoglobin concentration of:

- ❖ **Females adults** **12-16 g/dl.**
- ❖ **Males adults** **13.5-17.5 g/dl.**
- ❖ **Infant** **14 - 20 g/dl.**

- Hb quantity above this range, the case called **Polycythemia.**
- Hb quantity lower this range, the case called **Anemia.**
- Hb quantity was higher in male than in women **because the RBCs count in male was higher.**

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Blood Cell Count

(Lab: 4)

Assistant lecturer: Amina J. Mohammed

Blood:

It consists of:

1. Cells [erythrocytes (RBC) , leukocytes (WBC) and platelets].
2. Plasma.

1- Red Blood Cell (RBC) count

Introduction and Principle:

The number of (RBCs) in a blood sample can be estimated by the very old and traditional method using Neubauer's counting chamber, this method is time consuming and unreliable. It might be practiced in few laboratories when there is no better option, or for teaching purposes.

Objectives

- At the end of this session the students should be able to count the RBCs of a blood sample and identify the difference between the two sexes regarding RBC count.
- At the end of this session the students should be able to identify the significance of this knowledge.

Materials and Instruments

1. The haemocytometer. The haemocytometer consists of:

a. Counting chamber:

Is a heavy glass slide, in the center of which is ruled platform. The platform is lower than the rest of the slide by 0.1 mm. contain **(2)** counting areas, each area contains on big nine square. The four corner squares are used for counting leukocytes and are divided into **16 smaller squares**. The **central square** is also divided into **25** squares each one of them is divided into **16** small squares. This means that the total number of these small squares inside the center square **$25 \times 16 = 400$ squares.**

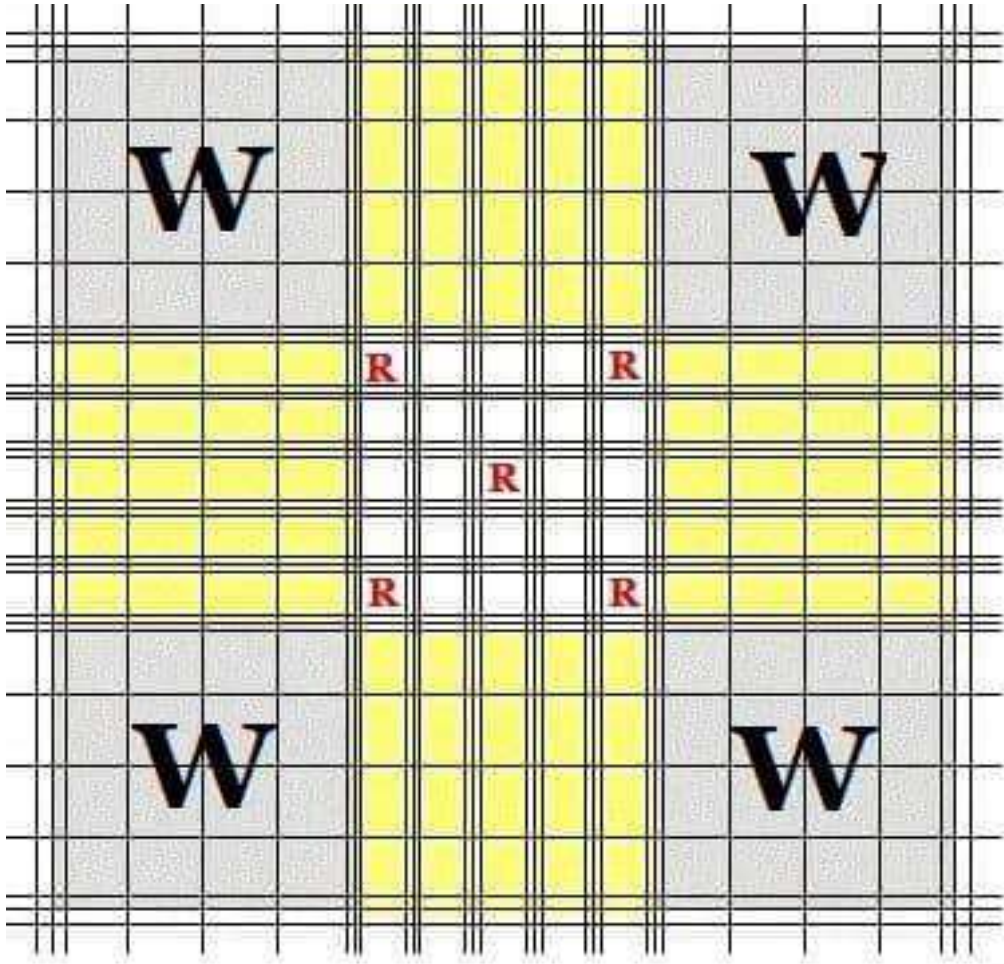


Figure (1): The ruled Neubauer's haemocytometer. (under microscope)



Figure (2): Haemocytometer counting chamber and cover glass.

The area of the big center square is **1mm^2** .

The area of the small square is **$1\ 400\ \text{mm}^2$** .

The thickness of the cover slide is **$1\ 10\ \text{mm}$** .

The volume of the small squares is **$1\ 400 \times 1\ 10 = 1\ 4000\ \text{mm}^3$** .

b. Special pipette (red cell pipette):

The pipette has a narrow stem, graduated with figures indicating 0.5 and 1, which widens into a bulb containing a red glass bead, which helps in mixing the blood with the diluting fluid (Hayem's solution). The bulb narrows again, and at this point, it is marked 101.

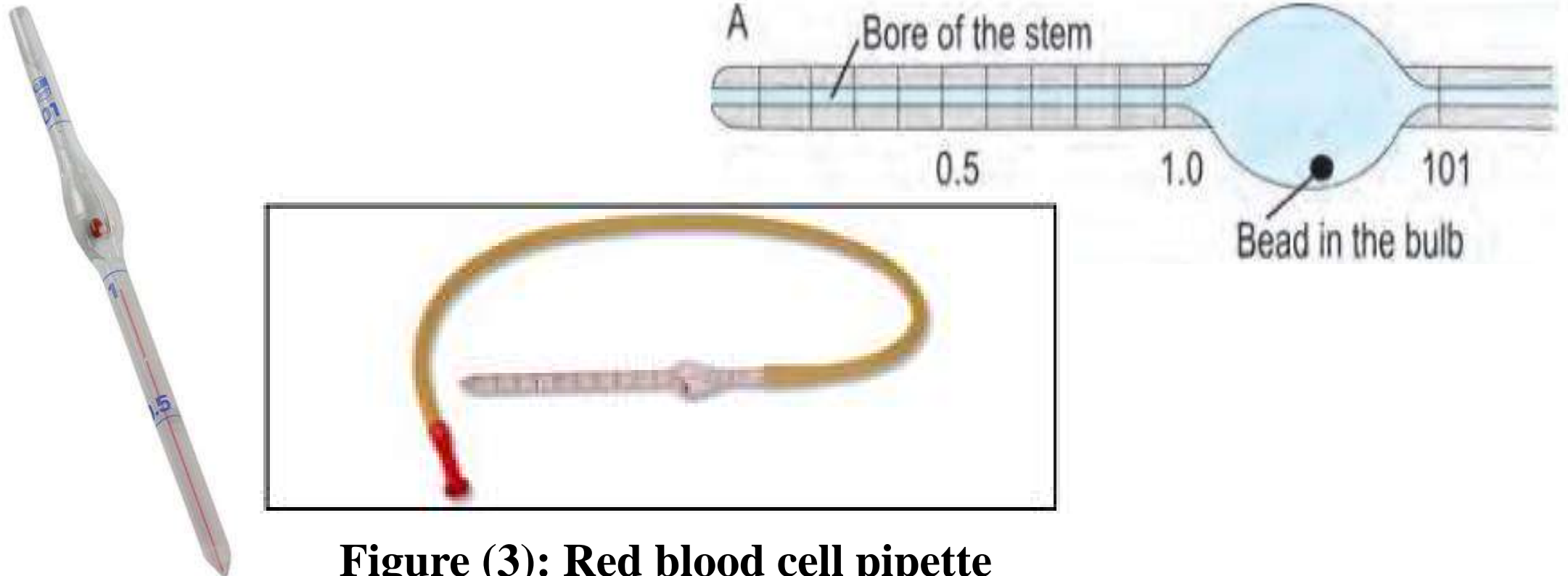


Figure (3): Red blood cell pipette

2. Hayem's solution (diluting fluid):

It consist of:

- sodium sulphate 10g.
- sodium chloride 2g.
- mercuric chloride 0.25g.
- distilled water 100ml.

3. Cover Slips.

4. Microscope.

5. Disposable lancets.

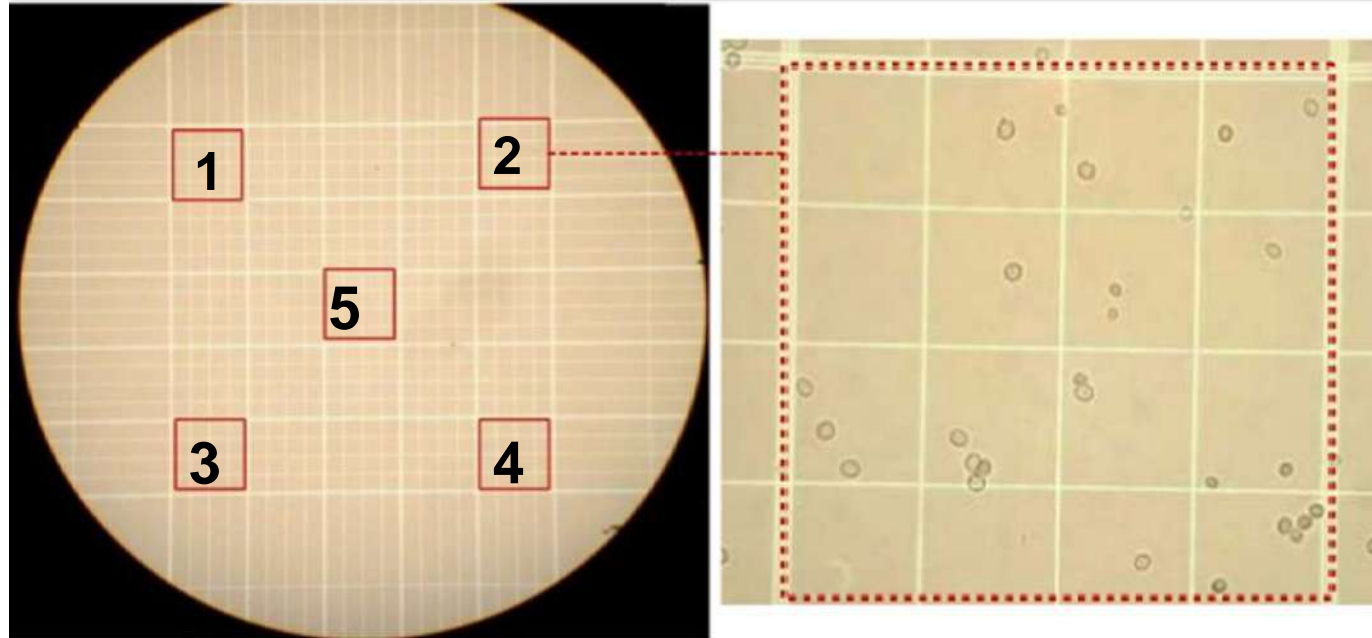
6. Alcohol 75%.

Procedure:

1. Wipe the finger-tip with an alcohol-damped swab and allow to dry in air (never blow).
2. Make a puncture on the finger's tip and draw blood up to the mark 0.5 with the RBC pipette.
3. Remove blood from the outside of the pipette with tissues.
4. Aspirate diluting fluid up to the mark 101 so that the dilution will be 1:200.
5. Hold the pipette horizontally between index and thumb of both hands and rotate gently for 1-2 minutes to ensure proper mixing.
6. After mixing discard the first three drops of the mixture.

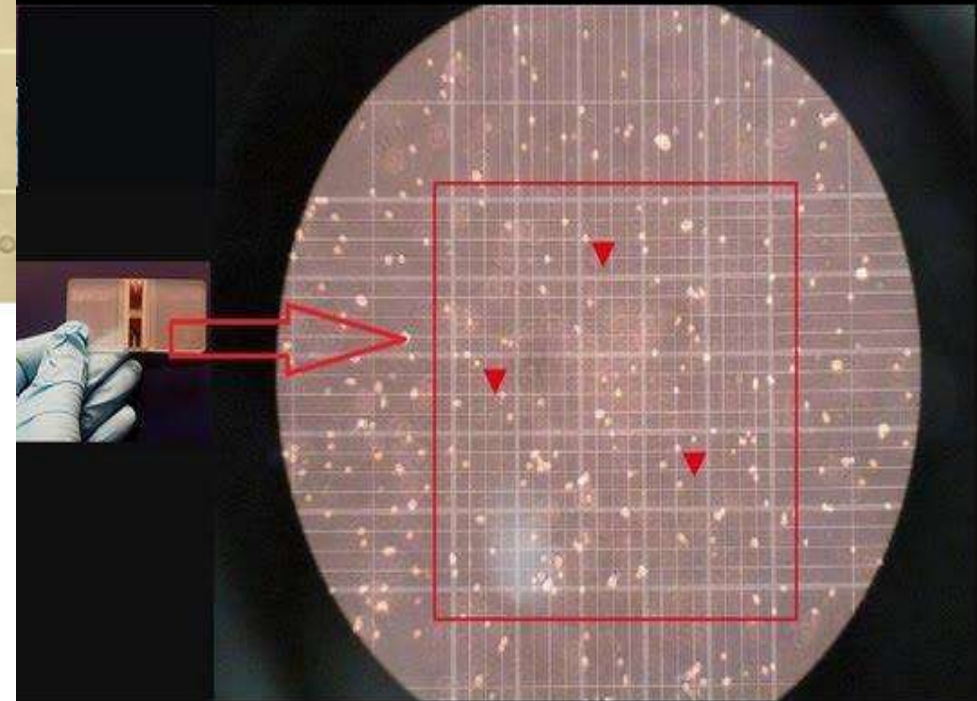
7. Fill the counting chamber with diluted blood by holding the pipette at **45°** with the counting chamber at the junction between the chamber and cover slip. The fluid will pass spontaneously on the chamber by capillary phenomenon (no need to blow).

8. Allow three minutes for the cells to settle down in the chamber before counting. Use the higher power **40X** objectives, start counting in the five designated squares.



Count all cells in specified squares, and multiply by the proper conversion factor; the number of cells per cubic millimeter can be determined.

Figure (4): Red blood cells counted in areas 1,2,3,4 and 5 of the Hemocytometer



Counting RBCs

Counting RBCs has done for **five squares** of the central square; the **total of them** is $5 \times 16 = 80$.

The **volume** of each of these squares is $1/4000 \text{ mm}^3$,

So the **volume** of the **80 square** = $1/4000 \times 80 = 1/50 \text{ mm}^3$.

The blood has been diluted **200 times**, so the numbers of RBCs in 1 mm^3 :

RBC count = $N \times 50 \times 200$.

RBC count = $N \times 10000$.

Normal range:

The RBCs range (**million/mm³** of blood):

- **Female: 3.8 - 5.8 million/mm³ of blood.**
- **Male: 4.5-6.6 million/mm³ of blood.**

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Packed cell volume (PCV)

(Hematocrit)

(Lab: 5)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

Is the volume of the red blood cells in 100 ml of whole blood, and expressed as a proportion of the total volume of the blood sample. The remaining volume of the sample is plasma, in addition, there is found a small amount of WBC and platelets _ measure as a percent.

Objectives:

At the end of this session the attendees will:

- ☐ Gain the knowledge and skills how to estimate packed cell volume.
- ☐ Indicate the significance abnormal PCV.

Methods:

There are (3) method for determination of hematocrit:

1. Microhematocrit method.
2. Macrohematocrit method (wintrobe method).
3. Electric method (coulter counter).

1. Microhematocrit method:

Principle:

Centrifuging the blood in special hematocrit capillary tube.

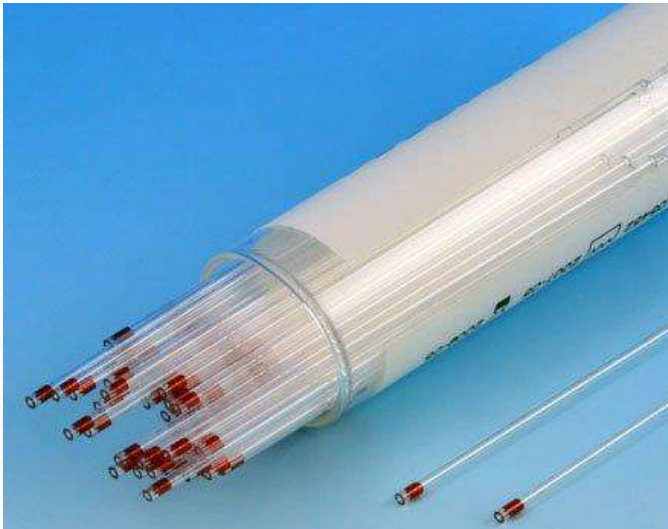
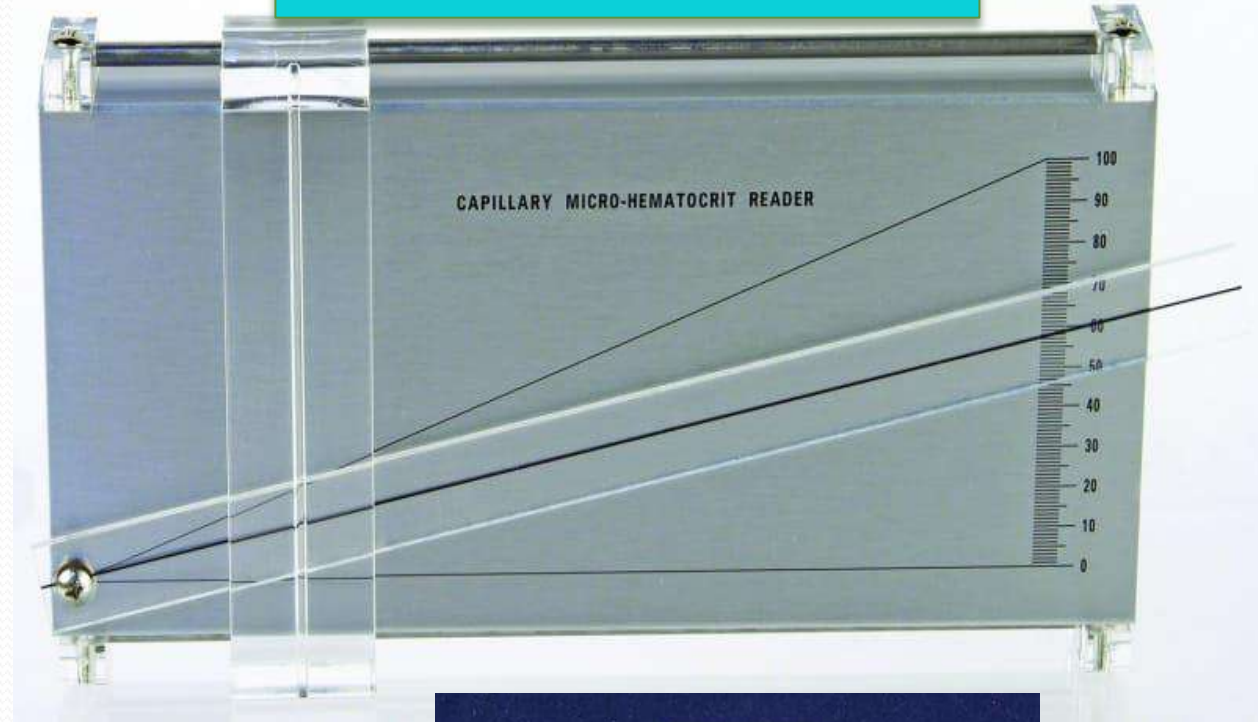
Materials and Instruments:

1. Microhematocrit centrifuge.
2. Microhematocrit capillary tubes (Heparinized).
3. Plastic seal to seal one end of the capillary tube.
4. Microhematocrit reader.
5. Cotton-wool, alcohol, and sterile lancets.

Microhematocrit centrifuge



Microhematocrit reader



Microhematocrit capillary tubes



Plastic seal

Procedure:

1. Blood is drawn into the tubes by capillary phenomenon by holding the tubes in a horizontal manner and allow $\frac{2}{3}$ to $\frac{3}{4}$ to be filled with blood. Air bubbles denote poor technique but do not affect the results of the test.
2. Seal the dry end of the tube by plastic seal or by heating the dry end of the tube rapidly on a fine flame of Bunsen burner combined with rotation.
3. The sealed tube is then placed in the radial grooves of the microhematocrit centrifuge with the sealed end away from the center of the centrifuge and centrifuged in 3000 rpm for 5 min.

1



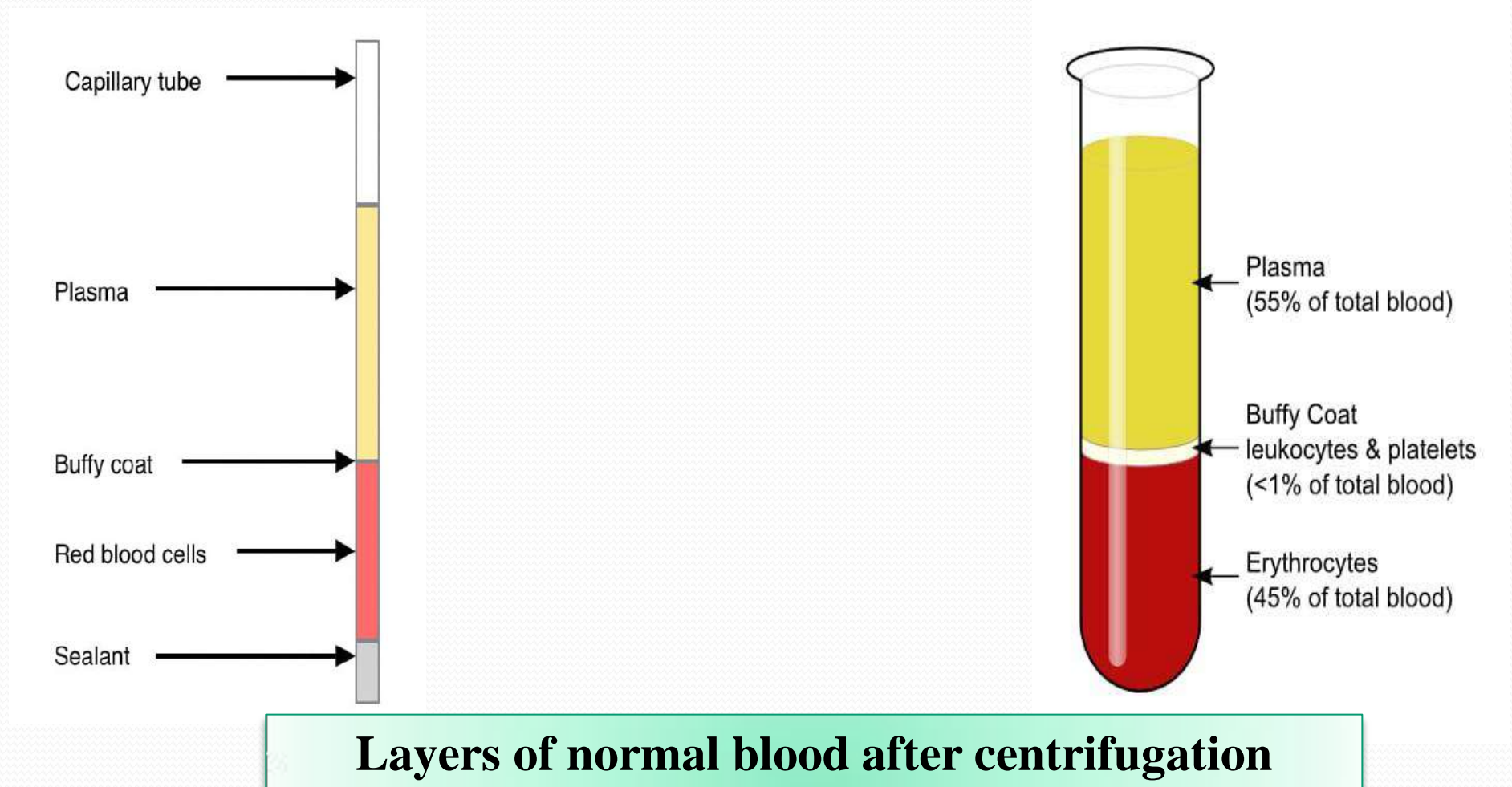
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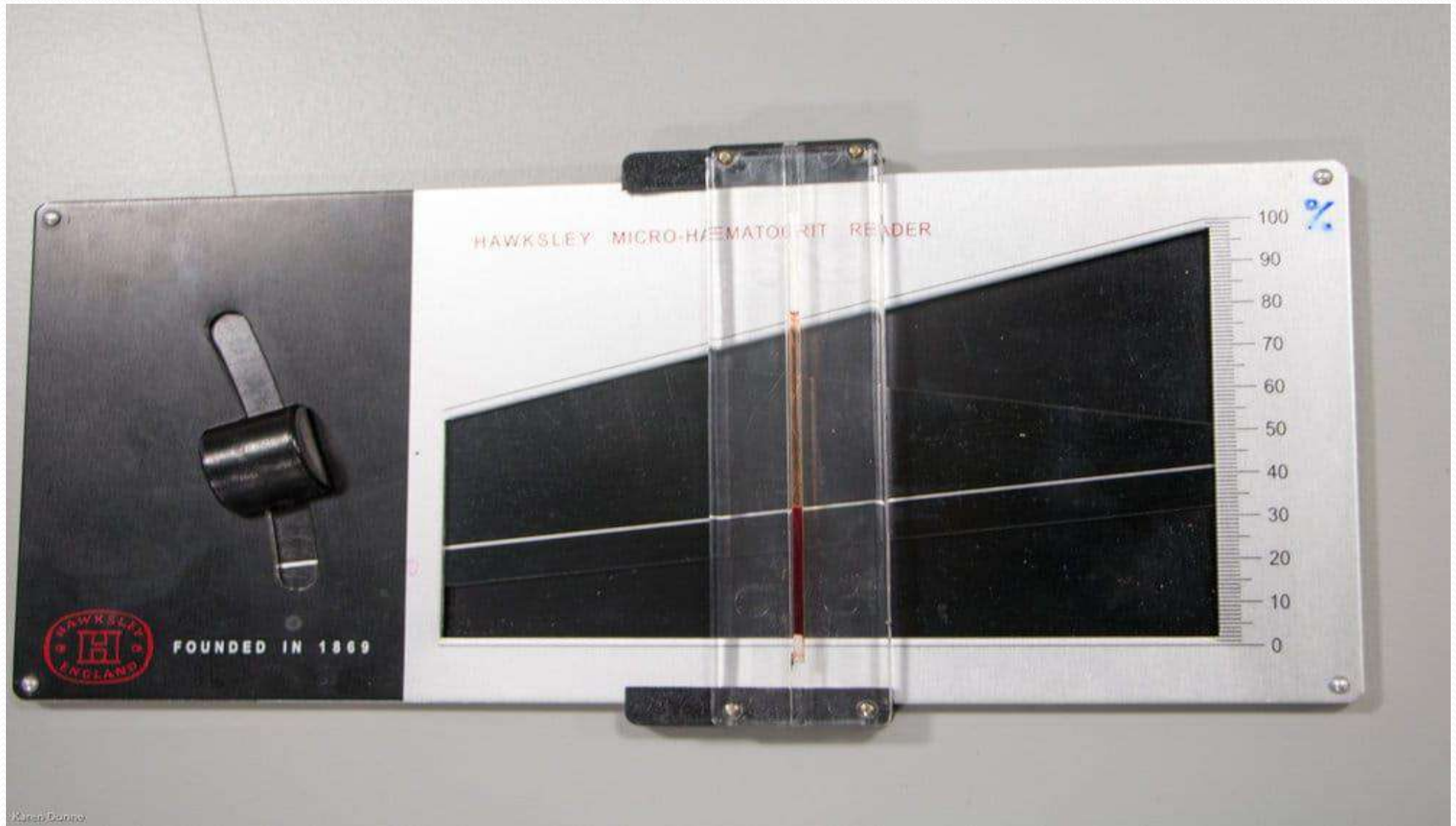
3



4. When looking at a centrifuged hematocrit tube, you can see three distinct layers. A top layer of clear slightly milky plasma, a thin buffy coat layer (consisting of W.B.Cs and platelets) and a dark packed R.B.C layer.



5. Obtain the results using the microhematocrit reader reading device, adjust the movable line to touch of the R.B.Cs in the tube as shown in the figure.



Results:

The normal range of packed cell volume is:

- **37-47% for females.**
- **40-54% for males.**

PCV is used:

- ❖ To determine anemia.
- ❖ In conjugation with the Hb concentration to calculate the mean corpuscular Hb concentration.

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Blood Cell Count

(Lab: 6)

Assistant lecturer: Amina J. Mohammed

2. White Blood Cells (WBC) Count

Introduction and Principle:

The white blood cells (Leukocytes) are considered as the mobile units of the body's protective system. They are responsible for combating different infections and toxic agents, that can sometimes cause lethal disease conditions as in pneumonia and typhoid fever.

They are formed mainly in the bone marrow (the granulocytes, monocytes and few lymphocytes and plasma cells) as well as in the lymphatic tissues. After formation they are transported into the blood stream to the different parts of the body (areas of infection) where they are needed.

Leucocytes:

They are commonly known as white blood cells. They are larger than erythrocytes, contain a nucleus and do not contain hemoglobin.

Mode of Functions of Leucocytes:

Phagocytosis: It is a process by which leucocytes engulf bacteria and foreign material.

2. Heparin production: Basophils produce heparin, which prevents intravascular clotting.

3. Antibodies production: Lymphocyte produce Ab, which combat Ag that enters to the body during viral infection.

Objectives:

At the end of this session the students will be able to do WBC count and to indicate the significance of the test in assessing patient's conditions.

Methods:

1. Manual methods.
2. Electronic Cell Counting (Coulter Counter).

The manual method of White Blood Cell Count

Materials and Instruments:

1. Whole blood, using EDTA or heparin as an anticoagulant. Capillary blood may be used.

2. Turk's diluting fluid:

Glacial acetic acid 3 ml.

to hemolyse RBC

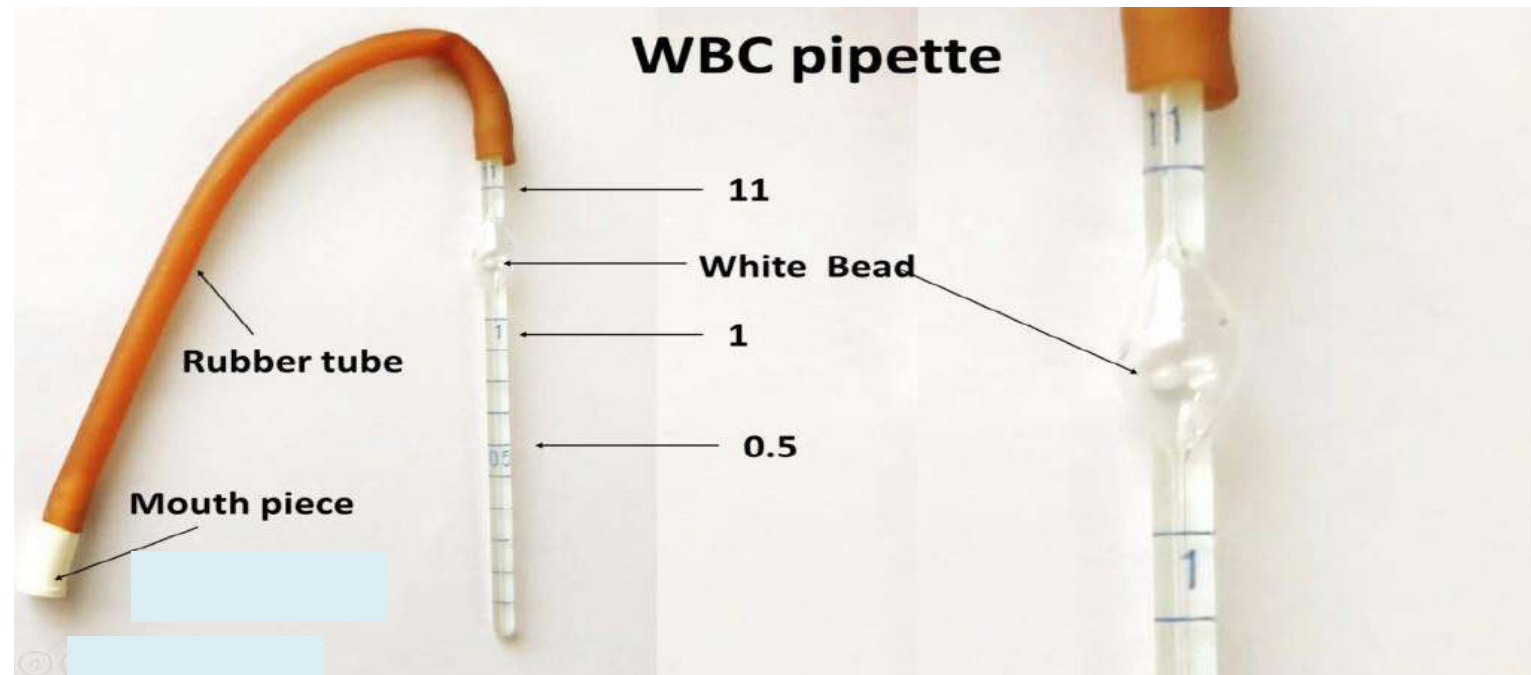
Gention violet (1%) 1 ml.

to color the nuclei of WBC

Distilled water 100 ml.

3. WBC pipette.

Figure : White cell pipette



4. Hemocytometer (Neubauer's counting chamber) with coverslip.



5. Microscope.

6. Lancets.

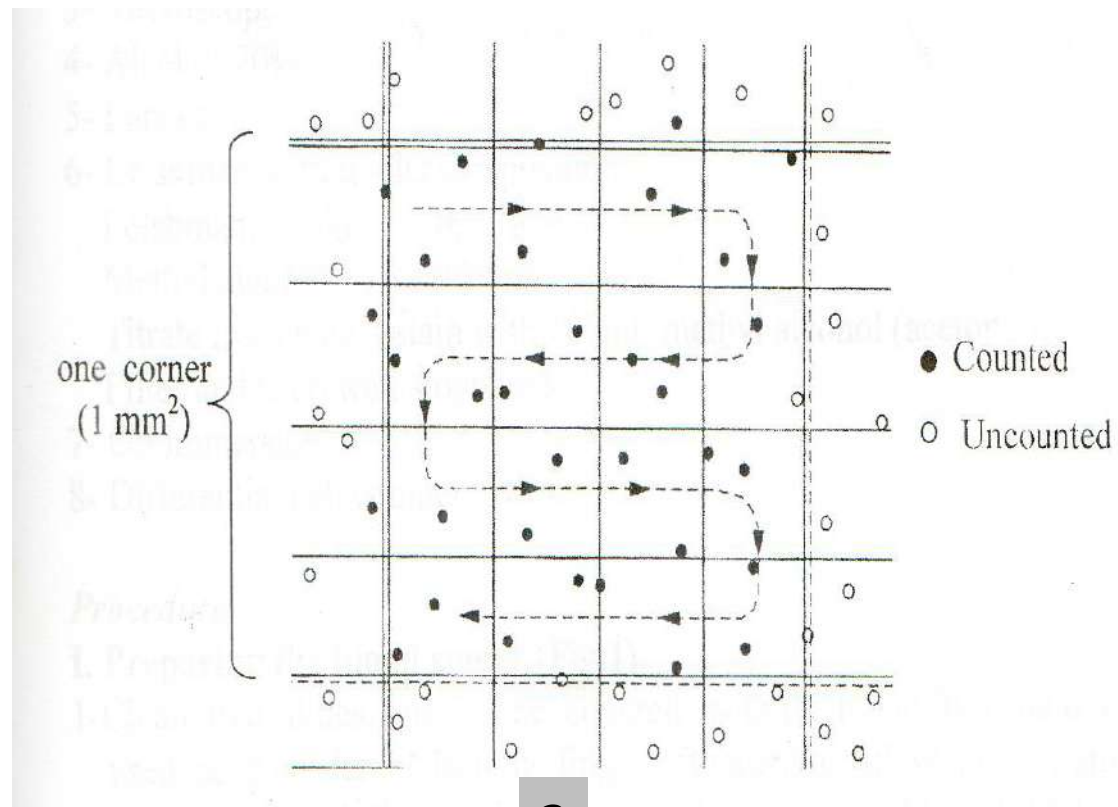
7. Alcohol 75%.

Procedure:

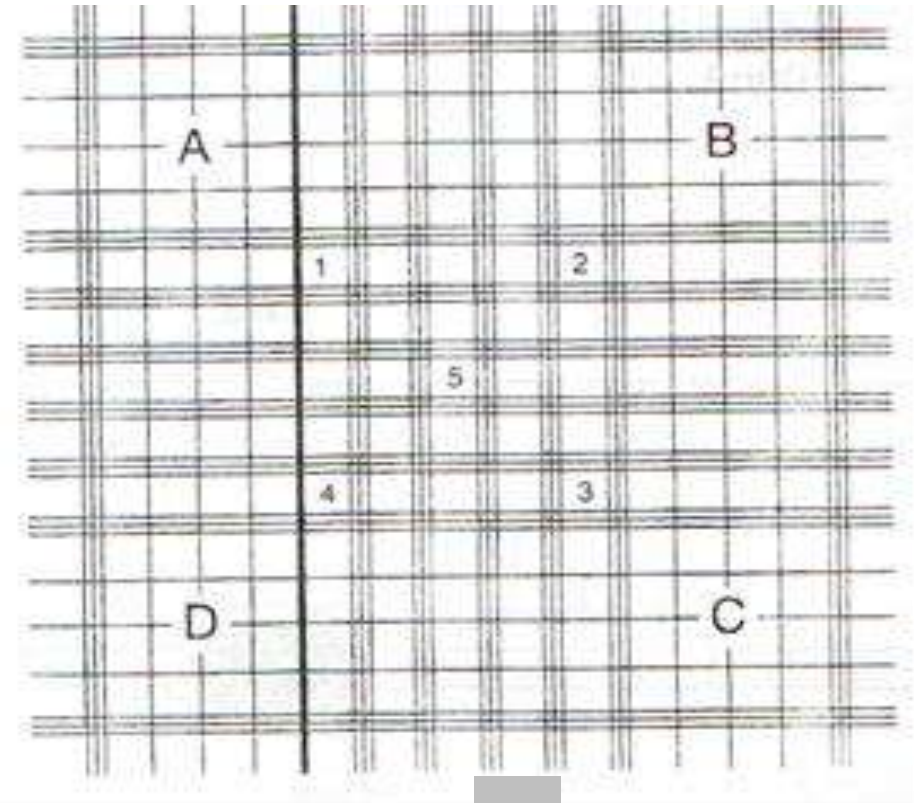
1. Obtain a drop of blood in the same manner as in RBC count. Draw blood up to the mark 0.5 using WBC pipette.
2. Aspirate diluting fluid up to mark 11. The dilution is **1 : 20**.
3. Remove blood from the outside of the pipette with a clean gauze.
4. Rotate the pipette horizontally with your hand for 3 minutes to ensure a proper mixing .
5. After mixing, discard the first drop of the mixture.
6. Fill the counting chamber with diluted blood by holding the pipette at 45° with the slide, and allow the mixture to seep under the coverslip. It should be allowed to stand for 1 minute prior to counting.

7. Use low power **10x** objectives first then pass to the higher power. Scan the four large corner squares of the chamber marked A, B, C and D as shown in the figure (1).

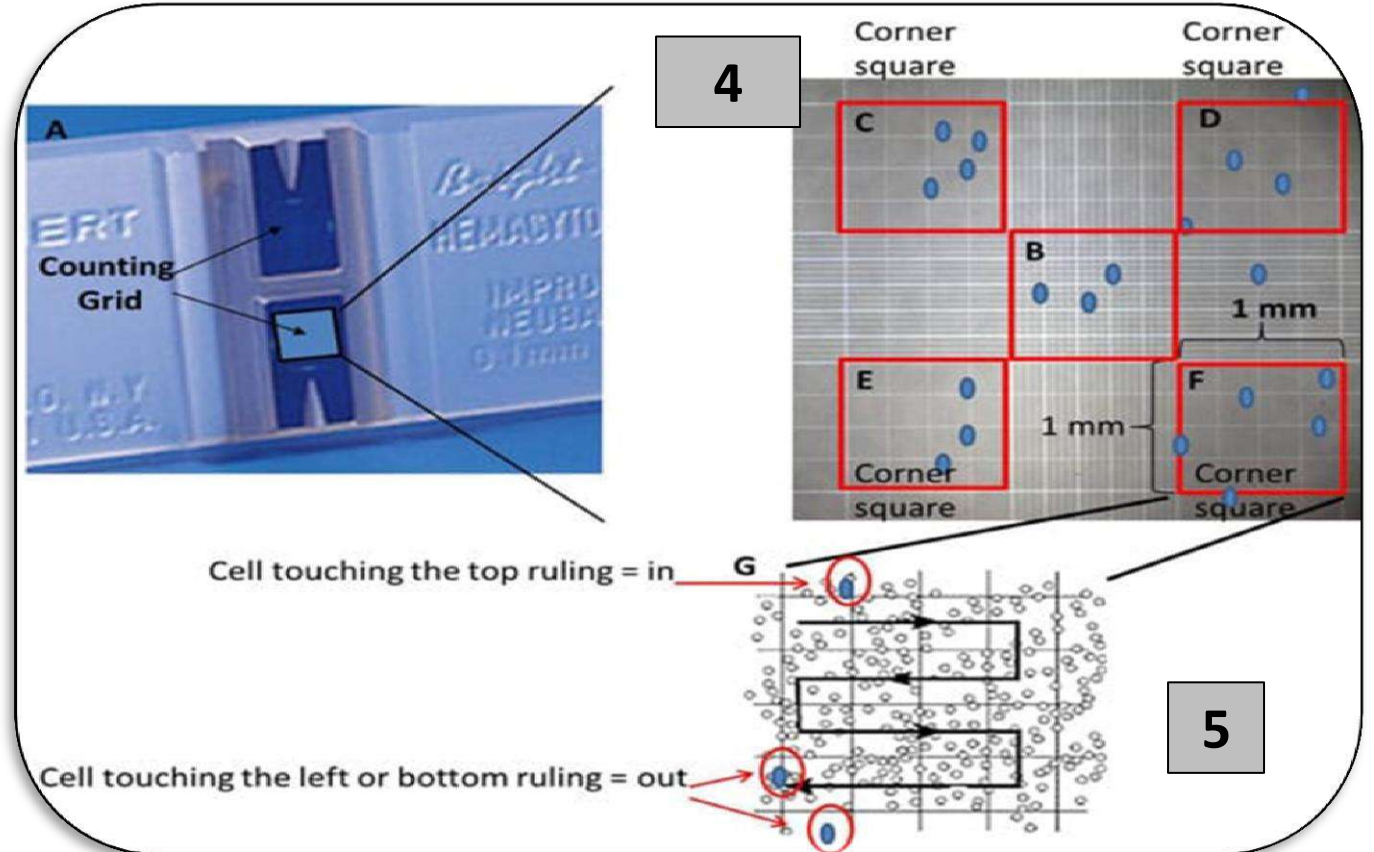
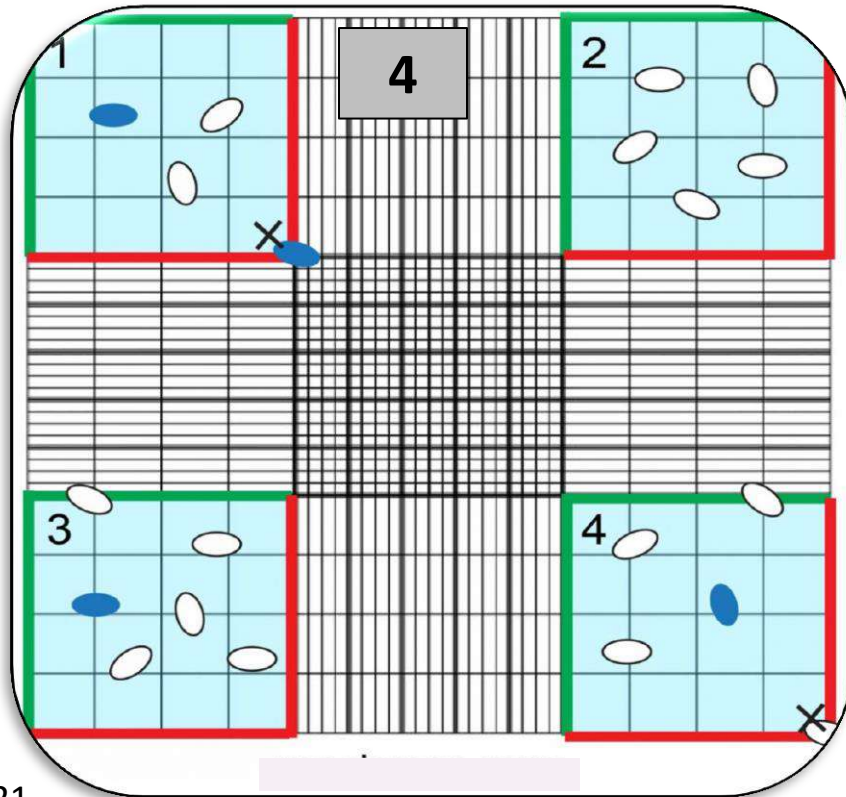
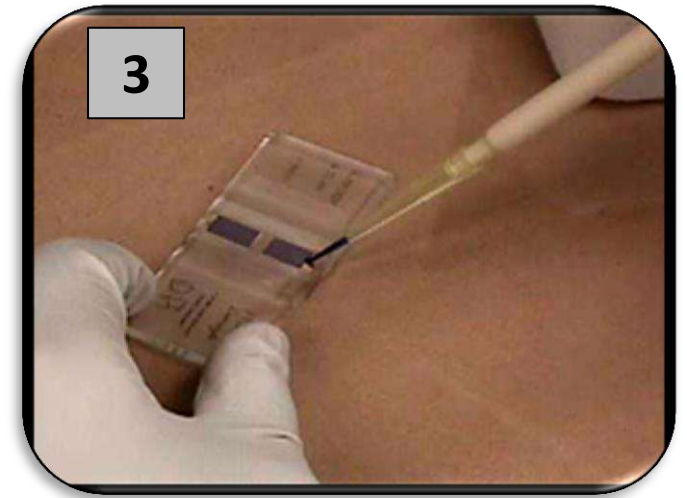
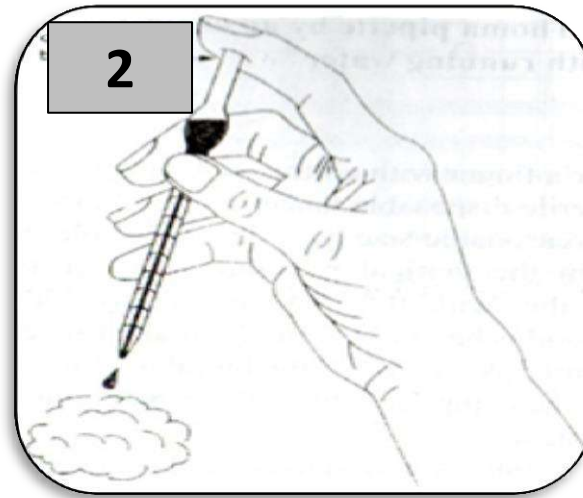
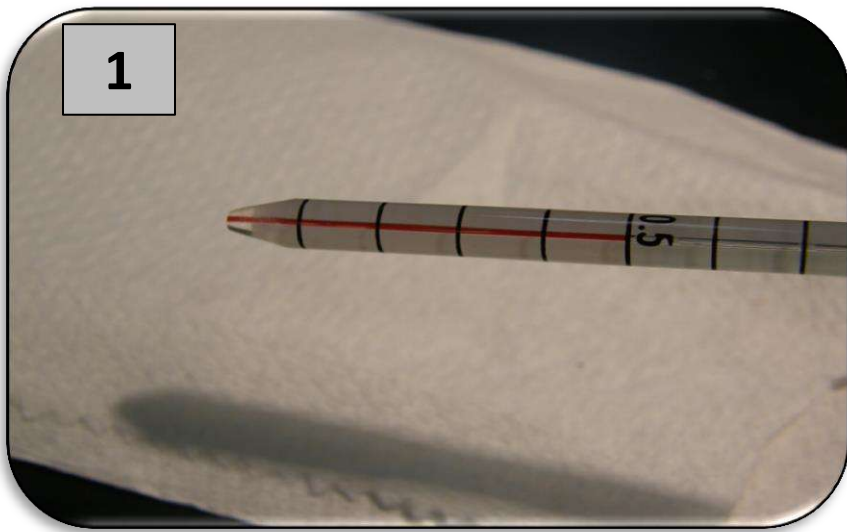
8. Count all cells that touch the left and upper outside lines, discharging those that touch the right and lower outside margin. The WBC look like black dots as shown in the figure (2).



2



1



Calculations:

- The volume of one square = $\frac{1}{10}$ c. mm.
- The volume of four squares = $4 \times 0.1 = 0.4$ c.mm.
- The no. (N) of WBC in 1 c.mm. = $N \times 2.5$.
- The dilution is 1:20
- Therefore, the no. (N) of WBC in a whole blood / c.mm. = number of WBC in four corner squares $\times 2.5 \times 20$.
- **Total WBC count / c.mm. = $N \times 50$**

Normal range:

Adult: 4000 -11000 cell /c.mm.

Pathological and physiological variation:

- ❖ **Leukocytosis:** This term is used to indicate an increase in the white cell count and is a common feature in most infections. A physiological increase in leucocytes is seen in:
 - Menstruation.
 - Pregnancy.
 - Muscular exercise.
- ❖ A reduction in white cell count is known **Leucopenia**; this condition is seen in:
 - Bone marrow suppression by drugs and x – ray radiation.
 - Pernicious anemia.
 - Infections such as typhoid and malaria.

- ❖ Leukocytosis is associated with the presence of premature white cell in the peripheral blood. This condition which may prove fatal is termed **Leukemia**.

DIFFERENCES BETWEEN RBC AND WBC PIPETTE

	RBC pipette	WBC pipette
1)	It has a red bead	It has a white bead
2)	It has graduations up to mark 101	It has graduations up to mark 11
3)	Size of bulb is larger	Size of bulb is smaller
4)	Size of lumen is smaller	Size of lumen is larger

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Differential White Blood Cell count ; Blood smear & Staining

(Lab: 7)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

Two main types of white blood cells are normally found in peripheral blood. **granulocyte** and include (**neutrophils, eosinophils, basophils**), all of these types have the polymorph nuclear, all have one feature in common, the granular appearance of the cytoplasm, for which reason they are called granulocytes. either second type is **non-granulocyte** and includes (**monocytes, lymphocytes**).

The **neutrophils** and **monocytes** protect the body against invading organisms mainly by ingesting them (phagocytosis). The **lymphocytes** produce antibodies against foreign antigens.

The main features of each type

- **Neutrophils:** has (3-5) lobed nucleus, small pink cytoplasmic, purple nucleus.
- **Eosinophil's:** usually bilobed, rarely (3) lobe nucleus, red-orange cytoplasmic, purple nucleus.
- **Basophils:** un segmented bilobed, rarely (3) lobe nucleus, deep blue or reddish-purple cytoplasmic granules, blue- black nucleus.
- **Monocyte:** the largest cells in normal blood nucleus kidney-shaped, large blue-gray cytoplasm (non-granular), blue or purple nucleus.
- **Lymphocyte:** round or oval nucleus, light blue cytoplasm (non- granular), deep blue or dark purple nucleus.

The differential WBC count is performed to determine the relative number of each type of WBC in the circulating blood.

In specific disease entity a particular white cell type may show increase in number in the blood e.g. **appendicitis**, **bacterial infections** cause increase in **neutrophils**, **allergy** and **parasitic infestation** cause increased **eosinophils**, **viral infection** cause increased **lymphocytes**. **monocytes** are increased in **monocytic leukemia**.

Objectives:

At the end of this session:

1. The students will be able to do the differential count of white blood cells of a provided blood sample.
2. Indicate the significance of the abnormal results.

Methods:



Manual Method

Materials and Instruments:

1. Whole blood sample with EDTA as anticoagulant.
2. Leishman's stain.

Leishman's stain **0.15 g.**

Methyl alcohol **100 ml**

Titrate Leishman's stain with **100 ml** methyl alcohol (acetone free) filter and keep well stopped.

3. Alcohol 70%.
4. Disposable lancet.
5. Microscope.
6. Oil immersion.

Procedure:

I. Preparation of blood smear:

1. Clean two slides, one to be **covered** with the blood film and the other to be used as **spreader**. Disinfect the finger with alcohol prep, allow to dry in air, make a puncture with disposable lancet to obtain a drop of blood. Make fine touch to the blood drop with one slide (only a small amount is required).
2. Place the edge of the other slide on the surface of the first slide just in front of the drop of blood at 45°.
3. Move the spreading slide slightly backward to make contact with the blood, move it forward slowly and steadily along the 1st slide to spread the blood on it.

4. Allow the film of blood to dry at room temperature (the blood smear should be air dried).

The slide is labeled immediately with the student's name.

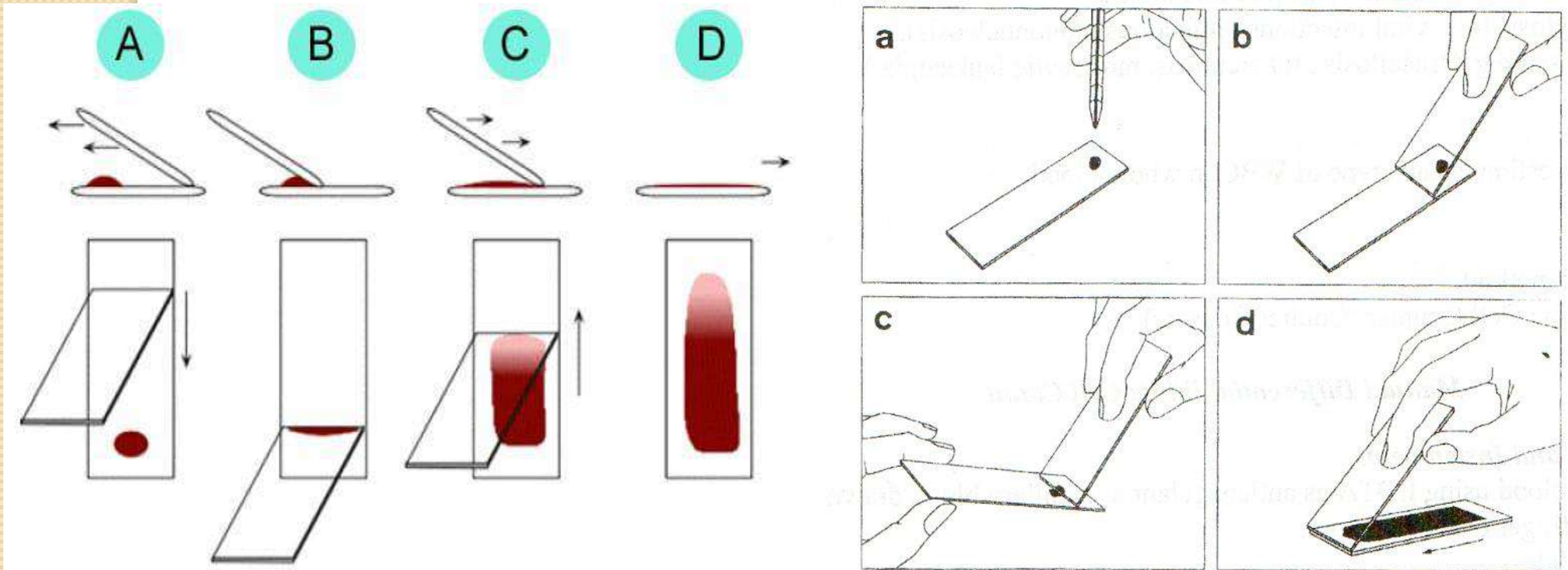


Figure (1): Preparing the blood smear.

II. Staining of blood smear:

1. Put the dried slide on a staining rack.
2. The blood smear should be stained as soon as possible, certainly within **1-2 hours**.
3. Carefully drop **Leishman's stain** on to the blood film until the film is covered completely.
4. Allow the stain to act for **1-2 minutes**.
5. Add distilled water to the stain, this gives dilution of **1:1** or **1:2**.
6. The diluted stain should act for **15-30 minutes**.
7. Wash the stain with distilled water, continue washing until the film look pinkish.
8. Shake off excess water and allow to dry at room temperature.



Figure (2): Staining of blood smear with Leishman's stain.

III. Examination of the stained blood film:

1. Place the slide (smear side up) on the stage of the microscope.
2. Examine the slide with the low power (**10×**) objective. Try to find an area where there are plenty of WBC, this area is usually located near the wedge shaped end of the smear.
3. Place a drop of **oil immersion** on the selected area and carefully change to the oil immersion objective lens (**100×**).
4. Count the different types of WBC, at the same time examine the morphology of the cells. Start counting in this area of the slide and gradually move the slide.

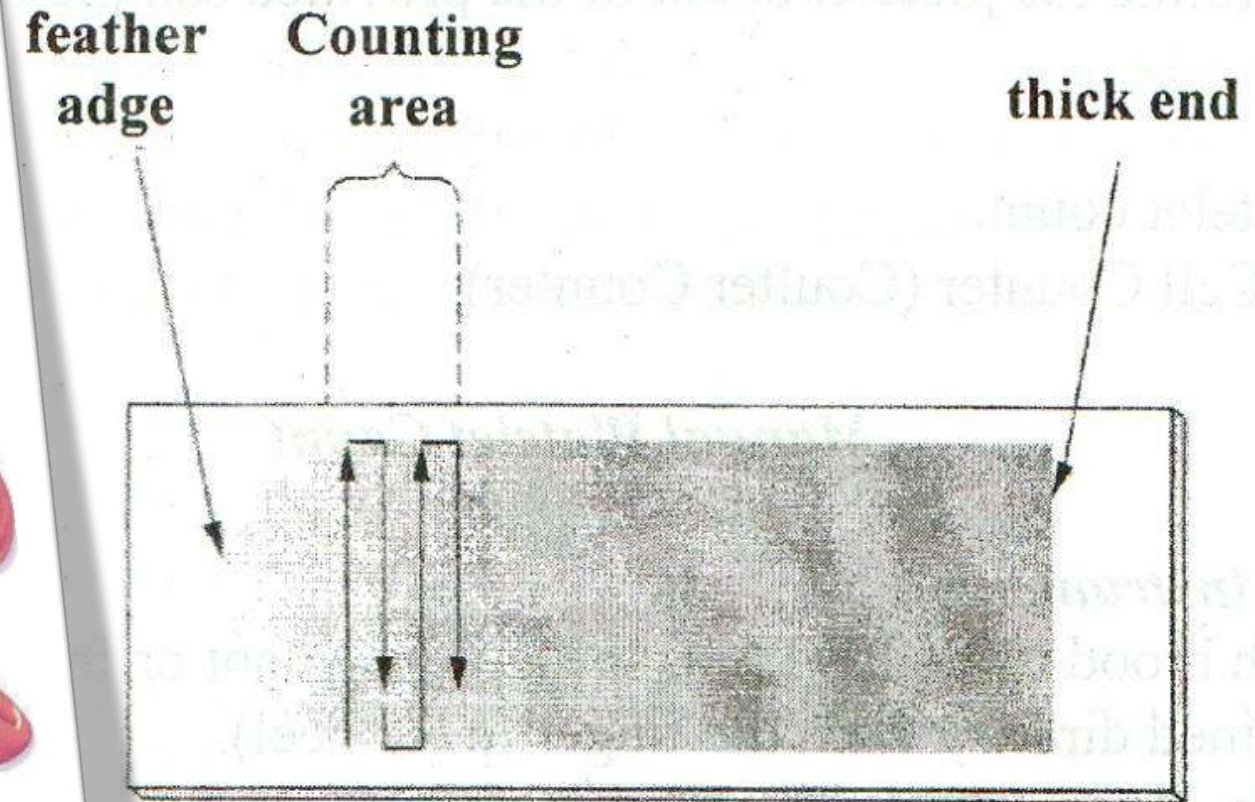
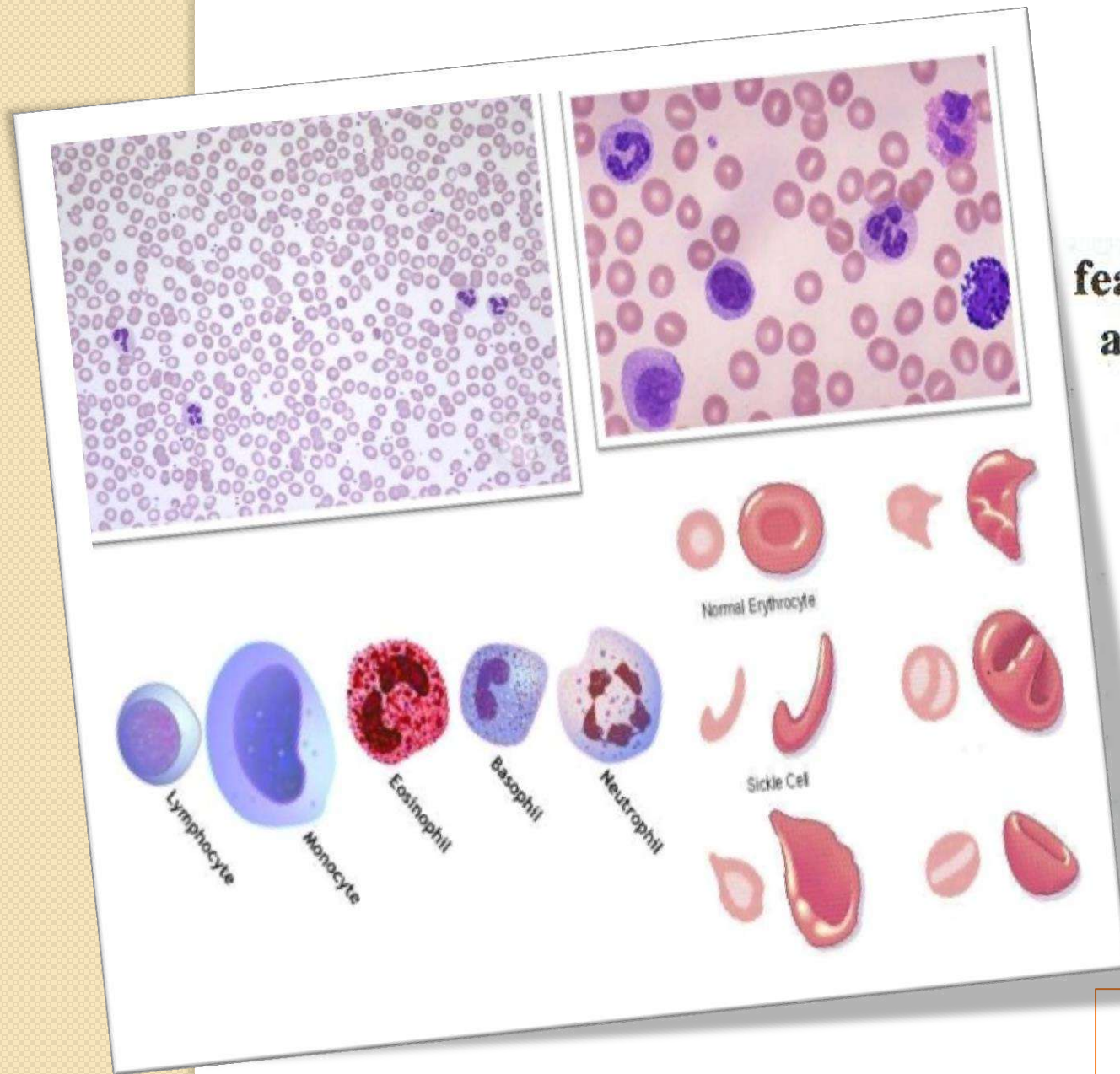


Figure (3): Blood smear showing a thin edge and the pathway for the differential cell count.

Calculations:

Count each WBC seen on the slide and record the number on a differential cell counter until **100 WBCs** are counted, then give the percentage of each type.

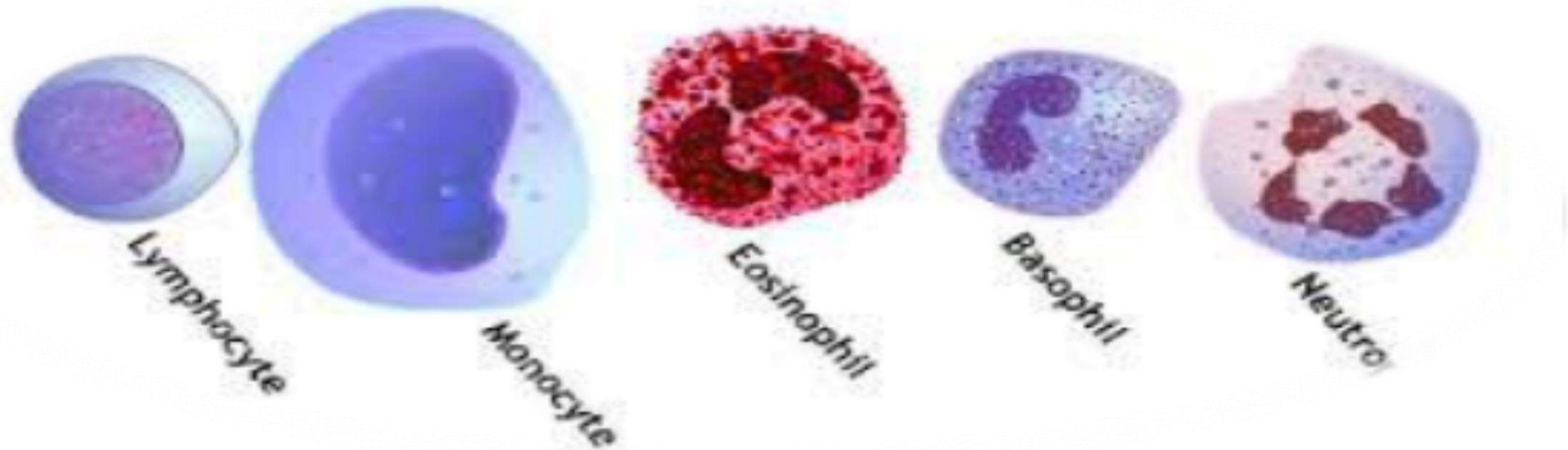


Figure (4) The different type of WBC with the different shape of nucleus and number of lobes of nucleus.

Results:

The normal values of the different types of WBC are as follows:

- ❖ Neutrophils 62% of total WBC.
- ❖ Basophils 0.4% of total WBC.
- ❖ Monocytes 5.3% of total WBC.
- ❖ Lymphocytes 30% of total WBC.

- ✓ **High neutrophil count (Neutrophilia):** an increase in number, seen in bacterial infection and inflammation e.g. tonsillitis, appendicitis.
- ✓ **Neutropenia:** a decrease in number, seen in typhoid , malaria.
- ✓ **High eosinophil's (Eosinophilia):** an increase in number, seen in allergy condition e.g. asthma and increase in parasitic infection.
- ✓ **Eosinopenia:** a decrease in number, seen in acute infection and steroid therapy.
- ✓ **Basophilia:** an increase in number, seen in chronic leukemia.
- ✓ **Lymphocytosis:** an increase in number, seen in viral infection.

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Platelet Count

(Lab: 8)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

Platelets are minute round or oval discs 2-4 micrometers in diameter. They are formed in the **bone marrow from megakaryocytes**, which are extremely large cells of the hemopoietic series in the bone marrow that fragment into platelets either in the bone marrow or soon after entering the circulating blood, especially as they try to squeeze through the pulmonary capillaries.

Despite the fact that they have no nuclei and cannot reproduce, they have many functional characteristics of whole cells, for instance, mitochondria and enzyme system that synthesize prostaglandins, remnants of both endoplasmic reticulum and Golgi apparatus, actin and myosin, fibrin stabilizing factor as well as growth factor that can cause vascular endothelial cells, vascular smooth muscle cells, and fibroblasts to multiply and grow, thus causing cellular growth that helps repair damaged vascular walls.

It has a half-life in the blood of **8-12 days**, at the end of which time its life processes run out.

Objectives:

At the end of this session the students will be able:

1. To determine the platelet count of a provided blood sample.
2. Indicate the significance of any abnormal result.

Methods

There are (2) methods for this count:

1. Electronic (Coulter counter).
2. Manual.

Manual method of platelet count:

Materials and Instruments:

1. Whole blood with EDTA as an anticoagulant.
2. WBC pipette.
3. pipette rotator.
4. Ammonium oxalate (1%) in distilled water, store in refrigerator and filter before use, for the hemolysis of the RBCs.
5. Microscope.

6. Petri dish.



7. Filter paper.



8. Hemocytometer used for RBC count.

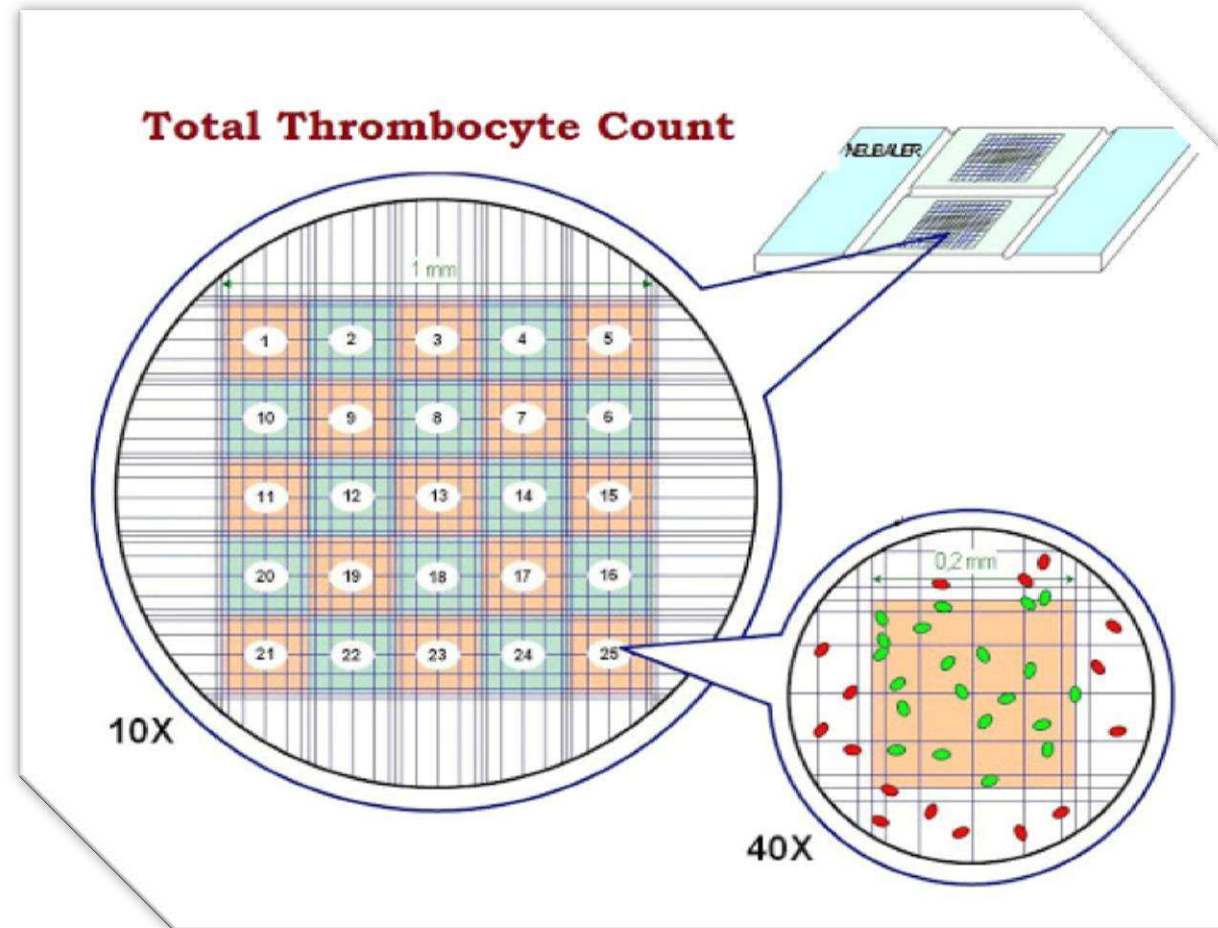
Procedure:

1. Using **WBC pipette**, with draw blood to exactly **0.5** mark and dilute to **11** mark with **1% ammonium oxalate (dilution 1:20)**.
2. Place the pipette on a pipette rotator for **10-15 minutes** to ensure complete hemolysis of RBCs.
3. Discard the **first three drops** of the fluid from the pipette and fill the counting chamber with diluted blood by holding the pipette at **45°** with the counting chamber at the junction between the chamber and cover slip. The fluid will pass spontaneously on the chamber by capillary phenomenon (no need to blow).

4. Place the counting chamber in **a moist petri dish containing** moistened filter paper with normal saline, leave the preparation to stand for **10-15 minutes** for platelets to settle down and prevents evaporation of the fluid is the counting chamber.

5. Carefully transfer the chamber to the stage of the microscope and examine under low power (**10×**). The background appears black with the white blood cells, platelets.

6. Change to high power (**40×**). The platelets appear as round or oval, with fine adjustment, The platelets are counted in the central square that is divided into 25 small squares , start counting in the **five** designated squares.



Calculations:

$$\text{Platelets/ c.mm} = \frac{N \times \text{dilution}}{\text{volume}}$$

Where N = average number of platelets per c.mm.

Dilution = 1:20

Counting platelets has done for **five squares** of the central square;
the total of them is **$5 \times 16 = 80$** .

The **volume** of each of these squares is **$1/4000 \text{ mm}^3$** ,
So the **volume** of the **80 square** = **$1/4000 \times 80 = 1/50 \text{ mm}^3$** .

$$\begin{aligned} \text{Platelets / c. mm} &= \frac{N \times 20}{\frac{1}{50}} = N \times 20 \times 50 \\ &= \mathbf{N \times 1000} \end{aligned}$$

Normal range:

- The normal concentration of platelets in the blood is between **150.000** and **300.000** per microliter (depending on the reference).

Homework

Questions and Discussion:

1. What is thrombocytopenia?
2. How would you differentiate between bleeding tendency due to thrombocytopenia from that caused by clotting factors deficiency?

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Erythrocyte Sedimentation Rate (ESR)

(Lab: 9)

Assistant lecturer: Amina J. Mohammed

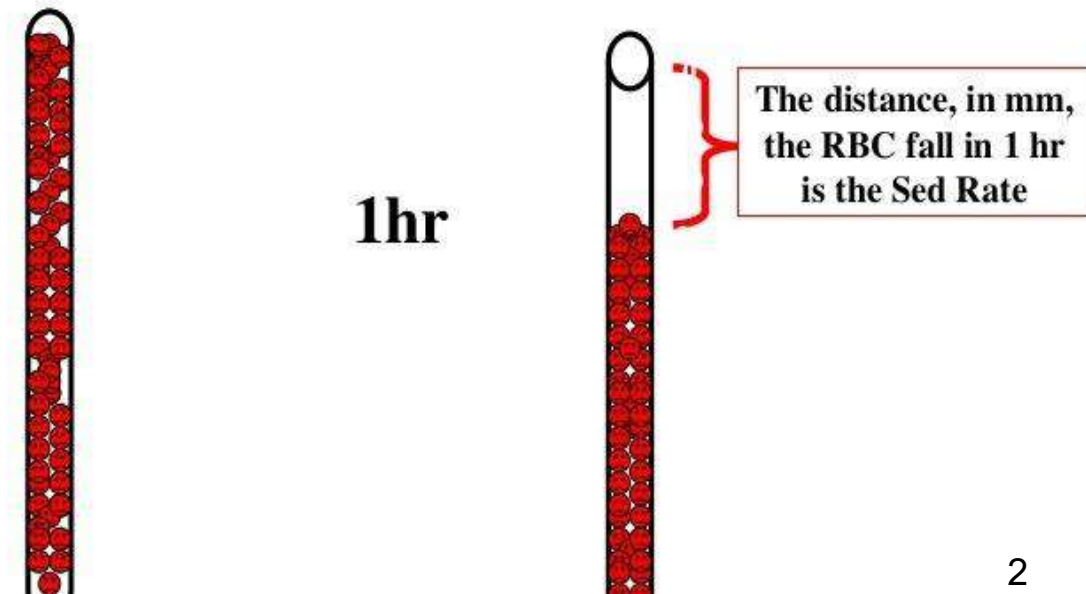
Introduction and Principle:

The determination of the rate at which the red cells settle or sediment (ESR) is often required by a physician to rule out the presence of organic disease or to follow the progress of a disease. ESR is generally done as part of complete blood tests.

The red blood cells of ant coagulated whole blood when allowed to stand for a period of time settle down from the plasma.

The distance (in millimeter) the red blood cells fall after a specific time period (1 hour) is known as the **Erythrocyte Sedimentation Rate (ESR)**.

Erythrocyte Sedimentation Rate (ESR)



Sedimentation of erythrocytes occurs through **three** phases:

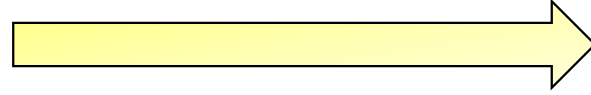
- 1- Red cells aggregates together and form rouleaux (clumping of red cells together like a stack of coins) and fall slightly.
- 2- The speed of falls is increased (Rapid settling of red cells).
- 3- The speed decreased during the third phase.

Objectives:

Upon completion of this session the students will be able to:

1. Estimate the ESR of a given blood sample.
2. Indicate the significance of the test.

Methods:



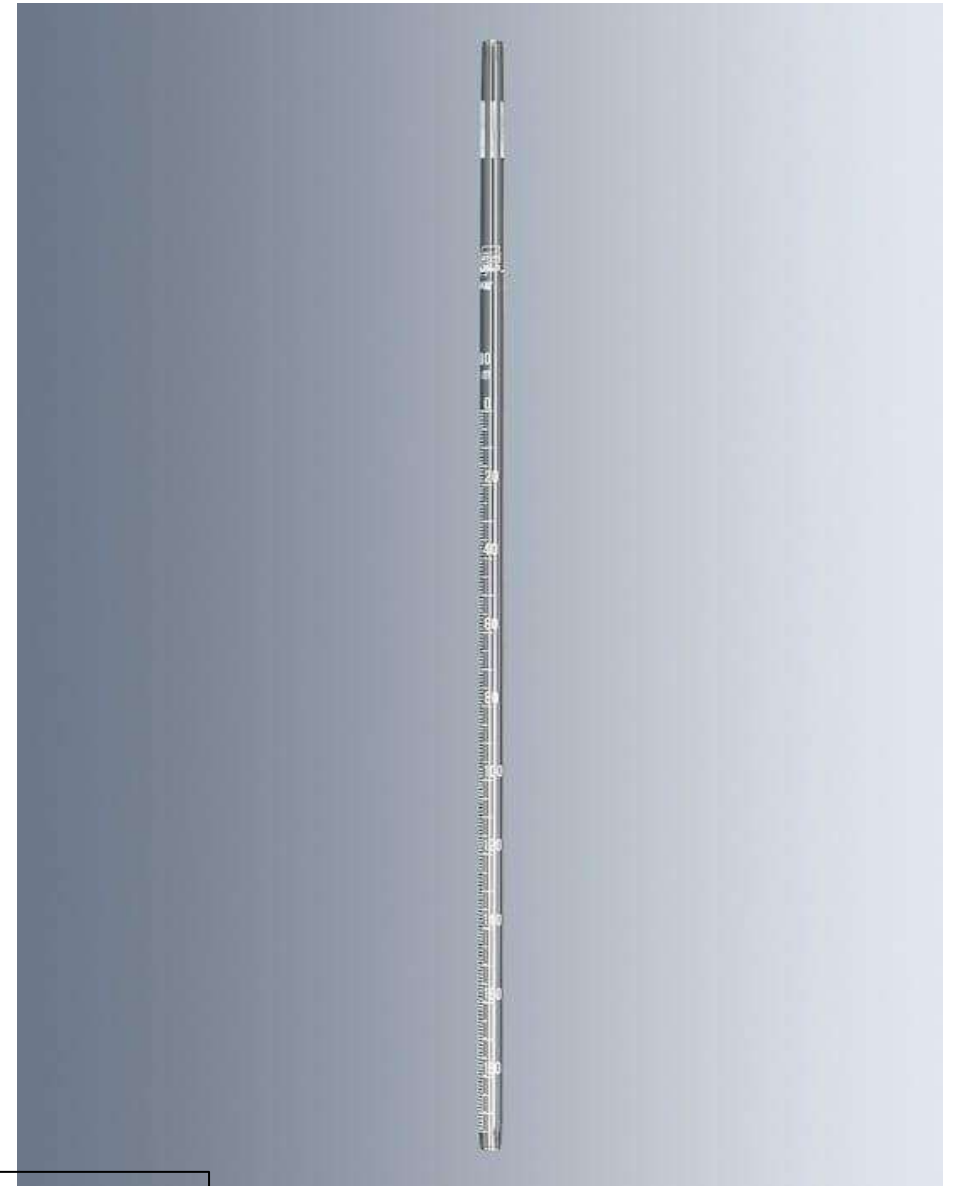
1. Westergren's method.
2. Wintrobe's method.

1. Westergren's method:

Materials and Instruments:

1. Sodium chloride 0.85% (W/V) as a diluting solution.
2. **Three ml** of whole anticoagulated blood. (EDTA as anticoagulant).
3. **Westergren's pipette** (A straight glass pipette **30 cm long** and **2.55 mm diameter**, graduated from **0-200** and open **at both ends**) (Fig.1).
4. Westergren's pipette rack (Fig.1).
5. Glass pipettes **2 ml** and **0.5 ml**.

6. Plain test tubes **13×100 mm**.
7. Disposable syringe, alcohol prep.
8. Applicator sticks.

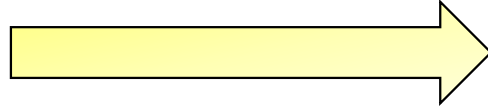


Procedure:

1. Mix the whole blood for at least **2 minutes** on a pipette rotator at room temperature
check the tube for clot formation using two applicator sticks.
2. Pipette **0.5 ml of 0.85%** sodium chloride in a plain test tube.
3. Add **2 ml** of well mixed whole blood to the test tube.
4. Mix the tube for **one minute**.
5. Fill the Westergren's pipette to exactly the **0 mark** making sure that there are no air bubbles in the blood column.

6. Place the tube exactly vertical into the Westergren's rack and leave it undisturbed for **60 minutes**, away from vibration and direct sunlight.
7. After **60 minutes**, read the number of millimeters the red blood cells have fallen (i. e. the height of the clear plasma above the upper limit of the column of the sedimented cells).
8. The result is expressed as **mm/hr**.

Results:



Normal range in adult males is **0-10 mm/hr.**

Normal range in adult females **0-15 mm/hr.**

2. Wintrobe's Method:

Materials and Instruments:

1. Wintrobe's tube 11cm length, 2.5mm internal diameter, graduated over 10cm scale. (Fig.2).
2. Wintrobe's pipette rack.
3. Disposable Pasteur pipette.
4. Whole anticoagulated blood.

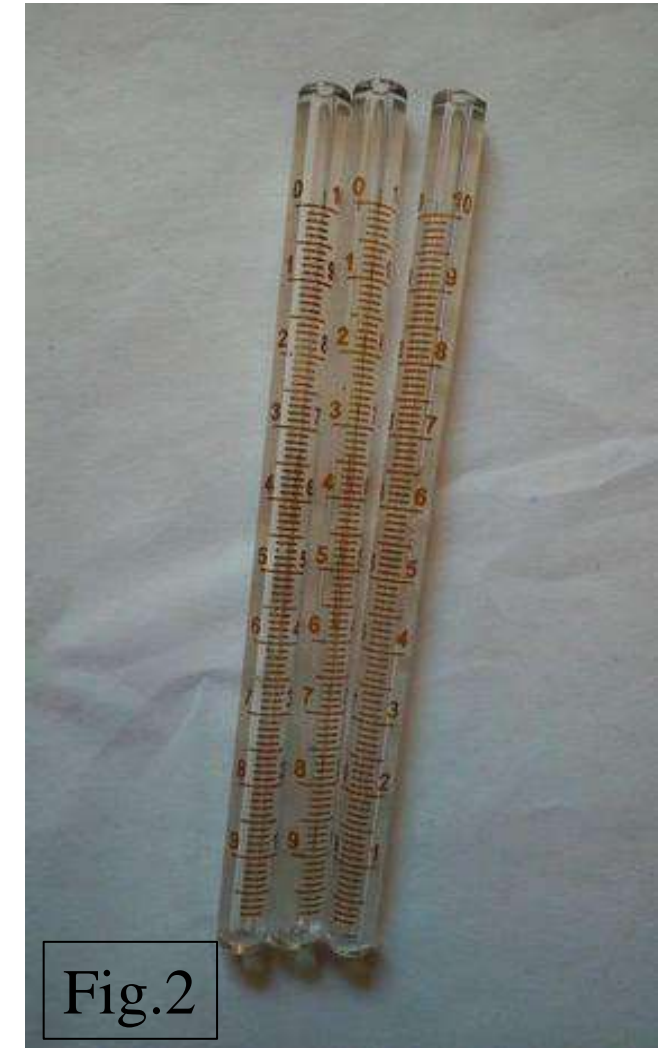
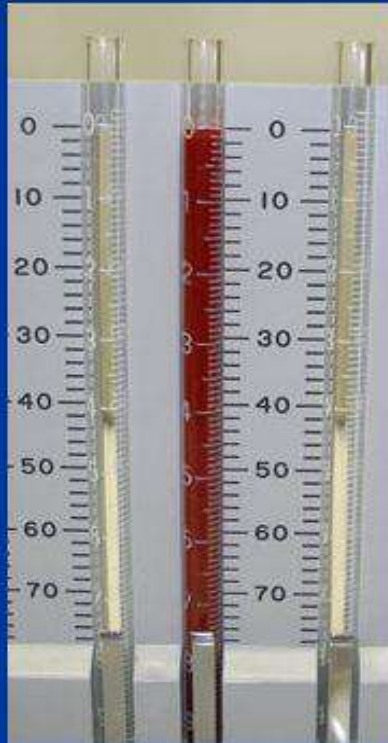


Fig.2

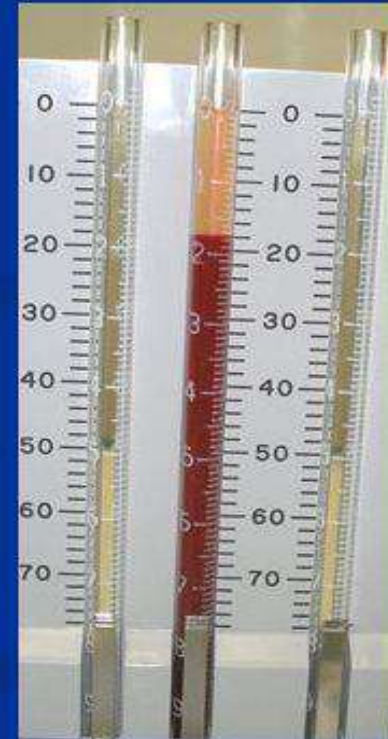
Procedure:

1. Mix the blood for at least two minutes on a rotator at room temperature.
2. With a Pasteur's pipette, fill the wintrobe's pipette to the **0 marks**.
3. Place the pipette in a vertical position in the rack.
4. Set the stopwatch for **60 minutes**.
5. After **60 minutes**, read the level of the clear plasma above the erythrocyte column.

ESR at zero time



ESR after one hour



Results:

- Normal range for men **0-9 mm/hr.**
- Normal range for females **0-20 mm/hr.**

Clinical application

E.S.R. increased in:

- 1- Rheumatoid arthritis.
- 2- Tuberculosis.
- 3- Acute and chronic infections.
- 4- Malignant diseases as myeloma.
- 5- In pregnancy and elderly people.
- 6- Anemia.

E.S.R. decreased in:

- 1- Polycythemia.
- 2- Congestive heart failure.

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Blood Groups & Rh. factor

(Lab: 10)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

The RBCs contain two main types of antigens known as **agglutinogens**, **type A** and **type B** are present on cell the surface of red blood cells membrane while the plasma contains antibodies anti-a and anti-b known as agglutinins.

To find out a person's blood group the RBCs are made to react with sera containing known **agglutinins**.

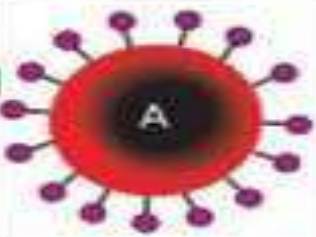
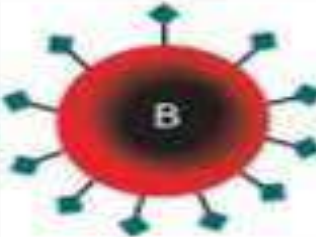








Accordingly blood groups are classified into **ABO blood groups**. Along with the ABO blood groups another main type of antigens (**antigen D**) may also be present on the red blood cells membrane accordingly blood types may be classified into **Rh groups**. At least 30 commonly occurring antigens and hundreds of other rare antigens have been found in human blood cells.

Each of which can at times causes the antigen-antibody reaction and there importance is principally for studying the inheritance of genes to establish parentage.

Because of the presence of the formerly mentioned subgroups blood transfusion may be mismatched and antigen antibody reaction ensues.

Therefore, it is a must to do another test before transfusing blood to exclude the presence of mismatched subgroups this test is called **'cross-matching test'** to determine ahead of time the presence of such subgroups that can create severe reactions.

Blood Type	Agglutinogens	Agglutinin	Frequency
O	-	Anti A and Anti B	47%
A	A	Anti B	41%
B	B	Anti A	9%
AB	A and B	-	3%

	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies present	 Anti-B	 Anti-A	None	 Anti-A and Anti-B
Antigens present	 A antigen	 B antigen	 A and B antigens	No antigens

Objectives:

Upon completion of this session the attendees will be able to:

1. Identify the types (ABO and Rh) of a provided blood samples.
2. Predict the hazards of transfusing mismatched blood.
3. Consider the precautions to be taken before blood transfusion.

Methods:



1. Slide method. ✓
2. Tube method.

Materials and Instruments:


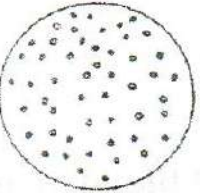
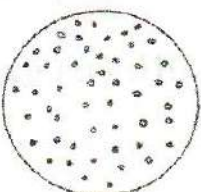



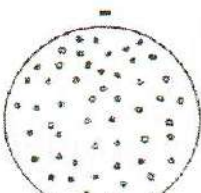
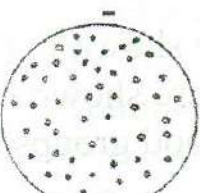
1. Anti sera (anti-A, anti-B, and anti-D). (**Fig:1**).
2. Slides.
3. Microscope.
4. Applicator sticks for mixing.



Fig (1): Antiserum A, B and D

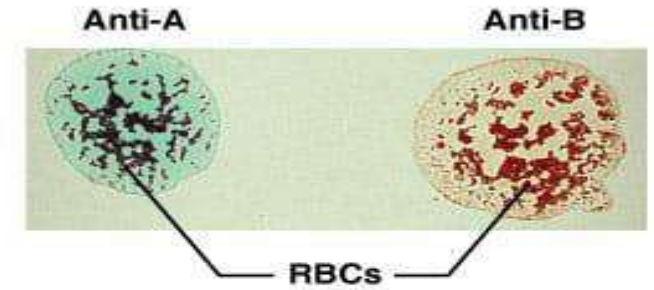
Procedure:

1. Place three drops of blood on the slide.
2. Then add one drop of each antibody (anti-A, anti-B, and anti-D) on each drop of blood.
3. Mix with an applicator stick, wait for three minutes, and read the result with the naked eye.
4. Examine under the microscope in case of confusion.
5. The ABO and Rh blood types are usually distinguished together as in figure (2).

Reaction of subject's cells to:		Blood group
Anti-A sera	Anti-B sera	
<div>+</div> 	<div>-</div> 	A
<div>-</div> 	<div>+</div> 	B
<div>+</div> 	<div>+</div> 	AB
<div>-</div> 	<div>-</div> 	O

Blood being tested

Type AB (contains agglutinogens A and B)



Type B (contains agglutinin B)



Type A (contains agglutinin A)



Type O (contains no agglutinogens)



Table (3): Rh (D) blood grouping.


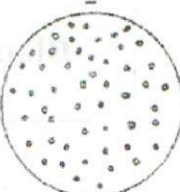
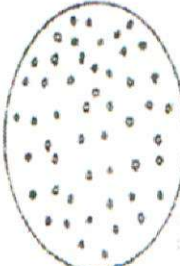

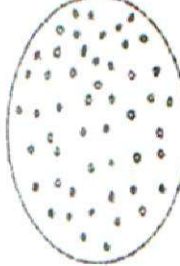
Reaction of subject's cells to:		Rh (D) group
Anti- D sera		
<div><div>+</div></div>		Rh ⁺
<div><div>-</div></div>		Rh ⁻

Table (4): Shows the ABO and RH blood groups.

Reaction of subject's cells to:			Rh (D) group
Anti- A sera	Anti- B sera	Anti- D sera	
-	+	-	B ⁻
<div style="text-align: center;">-</div> 	<div style="text-align: center;">+</div> 	<div style="text-align: center;">-</div> 	

Homework

What is the difference between agglutination and coagulation?

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Cross – Matching Test

(Lab: 11)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

Because of the presence of the subgroups in the blood of the donor and recipient, consequently it is not enough to typify the blood for safe blood transfusion.

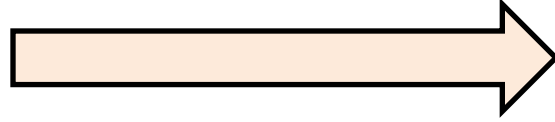
Therefore, **cross-matching** is considered as the direct test of compatibility of donor's cells and recipient's serum or plasma.

Objectives:

Upon completion of this session the attendees will be able to:

- ❖ Do cross matching test.
- ❖ Decide whether a donor's blood is compatible with recipient's blood or not?
- ❖ Predict the hazards of transfusing mismatched blood.

Methods:



1. Slide Method:

- 1. Slide method.**
- 2. Tube method**

Materials and Instruments:

1. Slides.
2. Serum or plasma from the recipient (containing antibodies).
3. Red blood cells from the donor (contain antigens)
4. Isotonic normal saline (0.9% NaCl) for dilution.
5. Applicator sticks.
6. Microscope.
7. WBC pipette.

Procedure:

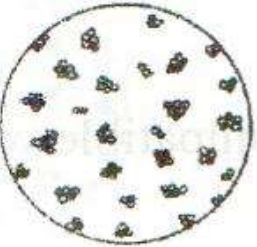
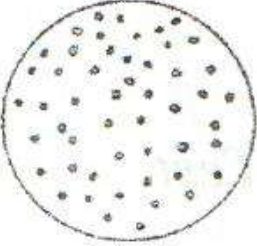
1. Prepare recipient serum or plasma.

Aspirate 2 ml of blood from the recipient and allow to clot (to obtain serum) or centrifuge with anticoagulant (to obtain plasma).

2. Donor's red blood cells from a finger's tip puncture should also be ready.

- i. Aspirate donor's red blood cells with the use of WBC pipette to the 0.5 mark, dilute with normal saline to the 11 mark.
- ii. Mix well and discard the first three drops.
- iii. Place one drop of diluted donor's red blood cells on a slide, add to it one drop of recipient's serum or plasma. Mix with applicator stick wait for 10 minutes.
- iv. Examine under the microscope.
- v. If blood is incompatible this will be shown as an agglutination or clumping of the red blood cells as shown **in the table (1)**.
- vi. Compatible blood will be shown as uniform suspension of RBC.

Table (1) : Shows blood compatibility (Cross Matching Test).

	Reaction between donor's cells and recepient's plasma antibodies	
a	<p>+</p> 	Incompatible
b	<p>-</p> 	Compatible

2. Tube method:

1. Blood sample from the recipient is centrifuged to obtain the serum.
2. The donor's red blood cells are washed by adding normal saline and centrifuged for three minutes to get rid of the plasma and all antibodies (agglutinins).
3. Label two (2) test tubes for each donor red cell suspension being tested with patient's serum or plasma.
4. Two (2) drops of patient's serum or plasma;
5. One (1) drops of the donor red cell suspension;

6. Mix the contents of the tube(s).
7. Incubate tube(s) at 37°C for 30 to 60 minutes.
8. Centrifuge tube(s). (Speed and time as recommended by manufacturer's instructions.)
9. Observe macroscopically for hemolysis and agglutination. Grade and record the results.
10. Wash the red cells three to four times with saline completely decant the final wash. Cell washer can also be used.
11. Add two (2) drops of AHG or IgG to the dry red cell button.
12. Centrifuge tube(s).

All samples should be checked with the naked eye and under the microscope. If agglutination takes place in the two tubes the donor's blood is considered as incompatible as shown in the figure (2).

Major Cross-Match

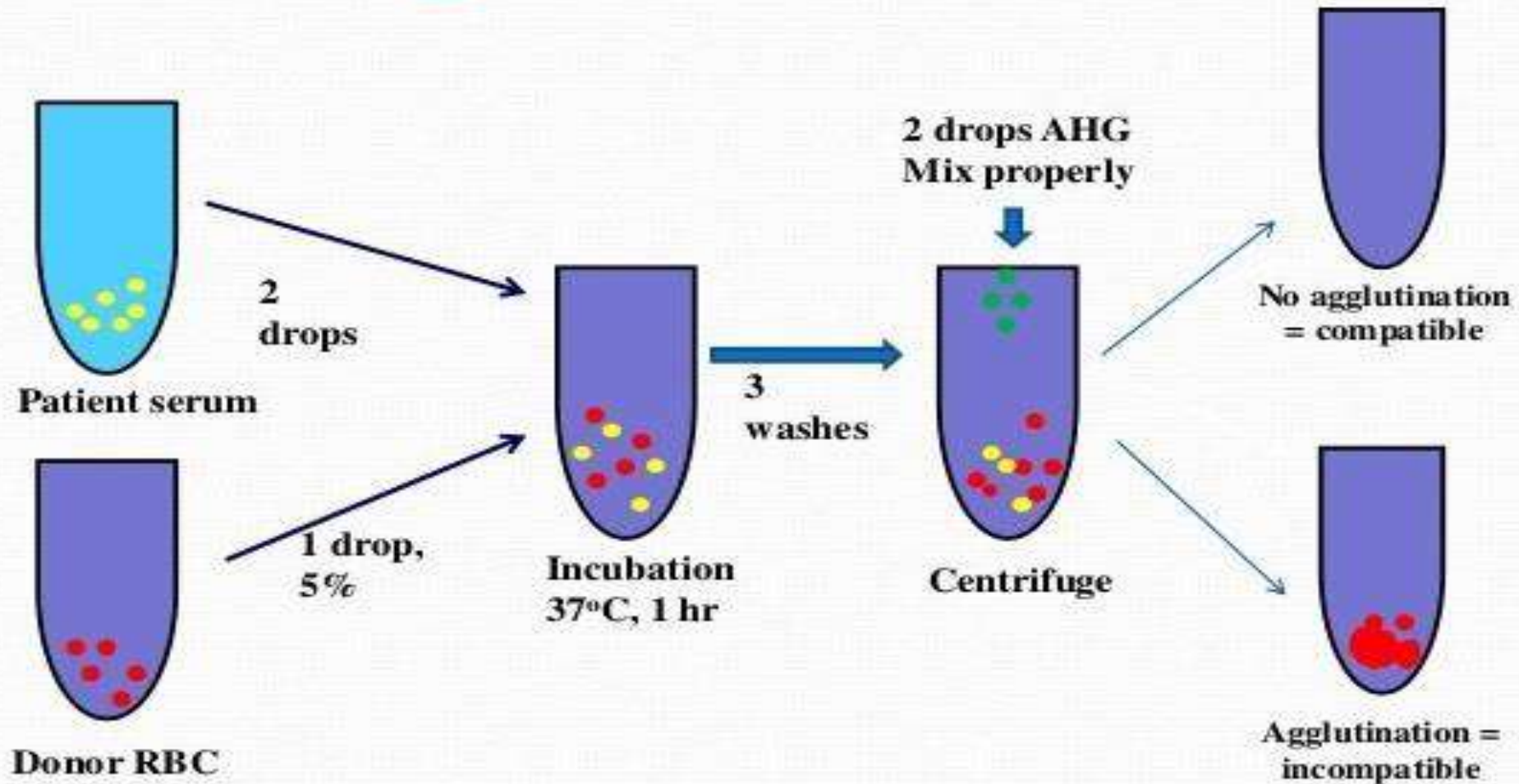


Figure (2): cross matching test (tube method)

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Hemostasis and Blood Coagulation

(Lab: 12)

Assistant lecturer: Amina J. Mohammed

Introduction and Principle:

The term **hemostasis** refers to the process of stoppage of bleeding after blood vessels are punctured, cut, or otherwise damaged.

Hemostasis means the bodily mechanisms responsible for preventing blood loss is a result of a complex, natural, physiological response.

Hemostasis involves the following (4) interrelated steps:

1. Vasoconstriction (contraction of injured blood vessels).
2. Platelet plug formation.
3. Formation of a blood clot.
4. Fibrinolysis (dissolution of the clot).

Physiological basis of bleeding:

Excessive and prolonged bleeding with small injuries or spontaneous bleeding may result from **defects of:**

- A. Platelets
- B. Blood vessel walls
- C. Coagulation of blood.

Bleeding time and clotting time

- **Bleeding Time (BT):** is the time interval between the skin puncture and spontaneous, unassisted (i.e .without pressure) stoppage of bleeding. The **BT** test is an in vitro test of platelet function.
- **Clotting time (CT):** is the time interval between the entry of blood into the glass capillary tube, or a syringe, and formation of fibrin threads .



Note

The BT and CT are two simple tests that are used as a routine before every minor and major surgery (e.g. tooth extraction), biopsy procedures, and before and during anticoagulant therapy, whether or not there is a history of bleeding.

I. Bleeding time:

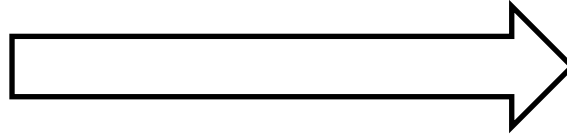
A standardized puncture of the ear lobe is made and the time course for bleeding to stop is recorded. Cessation of bleeding indicates the formation of platelet plugs which in turn depends on an adequate number of blood platelets and the ability of these platelets to adhere to each other and to the exposed collagen in the vessel wall to form aggregates.

Objectives:

At the end of this session the students must be able to:

1. Determine the time that elapses between the puncture of the skin and cessation of blood oozing (bleeding time).
2. Indicate the significance of abnormal bleeding time.

Methods:



Duke test.

Materials and Instruments:

1. Sterile disposable lancets.
2. Stopwatch.
3. Filter paper or tissue paper.
4. Alcohol prep pads.

Procedure:

1. Clean the ear lobe with alcohol prep and allow to dry.
2. A standardized puncture (3 mm. deep) of the ear lobe is then made with the use of a sterile disposable lancet.
3. The stopwatch is started at the moment of the puncture.
4. Absorb the emerging blood with a filter paper or tissue. Don't apply pressure on the ear lobe.

5. In the first two minutes absorption should be repeated every 30 seconds, after that absorb every 15 seconds.

6. When bleeding ceases the stopwatch is halted and the bleeding time is estimated.

❖ Normally bleeding time vary from 1-5 minutes.

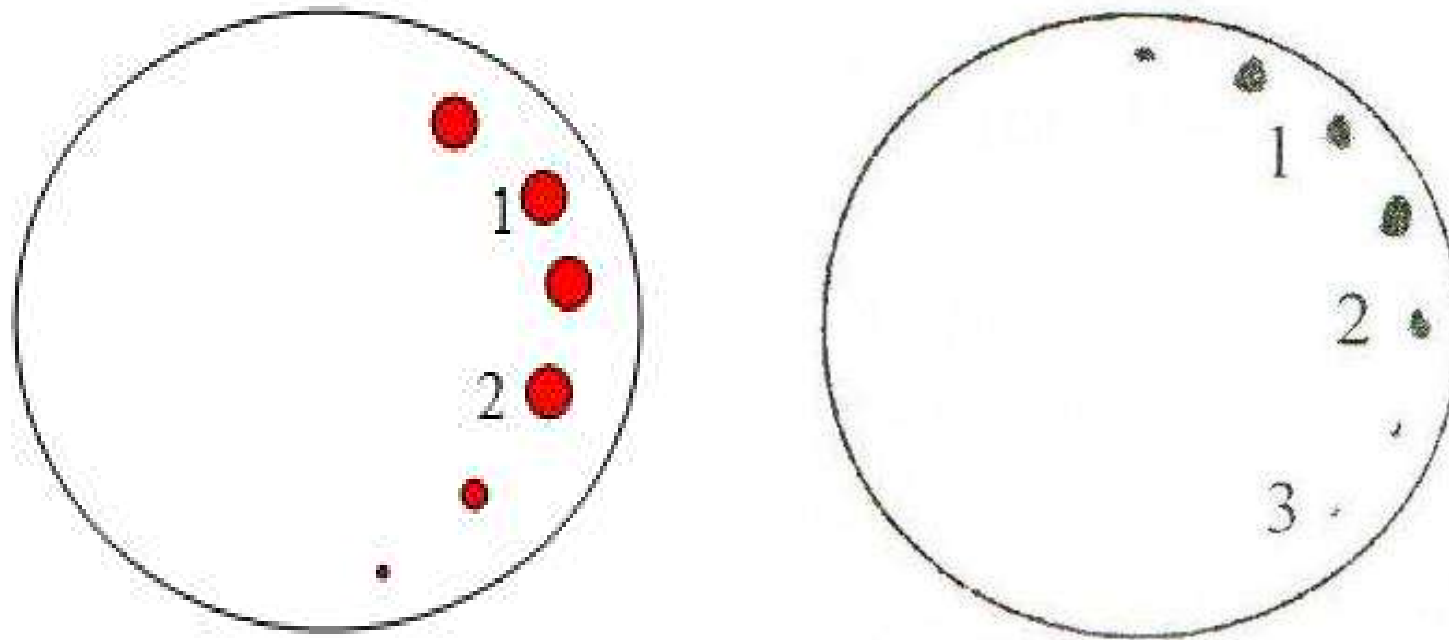


Figure (1): Normal blood spots on filter paper.

Homework

What is the clinical application
of determining bleeding time?

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Hemostasis and Blood Coagulation

(Lab: 13)

Assistant lecturer: Amina J. Mohammed

II. Clotting (coagulation) time:

The third mechanism for homeostasis is the formation of blood clot. The clot begins to develop **within 15 to 20 seconds** if the trauma of the vascular wall is minor and **after 1-2 minutes** if the trauma is major. Activator substances both from the traumatized vascular wall and from platelets and blood proteins adhering to the injured blood vessel wall initiate the clotting process.

Within 3 to 6 minutes after rupture of a vessel, if the vessel opening is not too large, the entire opening or broken end of the vessel is filled with clot. After 20 minute to one hour the clot retracts, this closes the vessel still further.

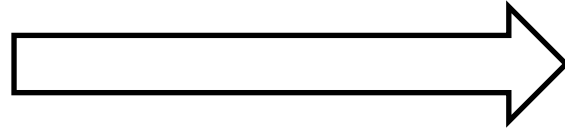
Therefore, the **coagulation time** of whole blood is the time required for a given amount of blood to clot under certain specialized condition. This process is depend on the blood clotting factors.

Objectives:

At the end of this session the students will be able to:

1. Determine clotting time.
2. Indicate the significance of abnormal results.

Methods:



1. Lee's and White method (test tube method).
2. Capillary method. ✓

1. Test tube method

Materials and Instruments:

1. Fresh whole blood 4 ml.
2. Sterile disposable syringe (10 ml) and 20-gauge needle.
3. Glass test tubes 13 × 100 mm.
4. Water bath 37 °C.
5. Stop-watch.

Procedure:

1. Label three test tubes with the patient's name and give them the numbers 1, 2 and 3.
2. With the use of disposable syringe withdraw 4 ml of venous blood.
3. Carefully place 1 ml of the blood in test tube 3, and start the stopwatch immediately. Then add **1 ml** of blood in **test tube 2**, and lastly, **1 ml** in **tube 1**.
4. Put the three tubes in a 37°C water bath.

5. At exactly **5 minutes**, tilt test tube 1 gently to 45° repeat this procedure every 30 seconds until the test tube completely be inverted without spilling the contents. i. e. the blood is completely clotted.
6. Record the time it took for the blood in test tube 1 to clot.
7. **Thirty seconds** after the blood in test tube 1 is clotted, proceed with tube 2 and repeat the same procedure, tilting this test tube every **30 seconds** until a clot is formed, record the results. Repeat the same procedure for test tube 3.
8. Since agitation and handling speed-up coagulation, the coagulation time of test tube 3 is handling the reported result. Normally clotting time ranges from **5-15 minutes**.

2. Capillary method

Materials and Instruments:

- ❖ capillary tube without heparin.
- ❖ lancet.
- ❖ stop watch.

Procedure:

1. Clean the finger tip with alcohol swab, and allow to dry. Make a puncture with sterile disposable lancet and allow a good drop of blood to accumulate.
2. Fill a capillary tube to it's full capacity you will need the full length of the tube because the tube is to be broken repeatedly until fibrin threads appear.
3. Put the capillary tube in the grove of the hematocrit reader and hold it in place with two finger-tips.
4. Cut the capillary tube into small pieces by pressing the tip of the cutter on the capillary tube between the fingers-tip, carefully separate the broken piece, using the tip of the cutter, by sliding the cut-off end of the capillary along the groove to see whether fibrin strands have formed or not.

5. The fibrin when formed appears as threads joining the two separate ends of the capillary tube.

6. For the first **two minutes**, break the tube every **30 seconds**, after that every **15 seconds**. The coagulation time is calculated from the time when blood appears from the puncture to the time when **fibrin is formed**. For accurate measurement, the temperature should be controlled.

Normal range of clotting time 2-8 minutes

Homework

What is the clinical significance of this test?

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Examination of the urine (Urine Analysis) (Lab: 14)

Assistant lecturer: Amina J. Mohammed

Objectives

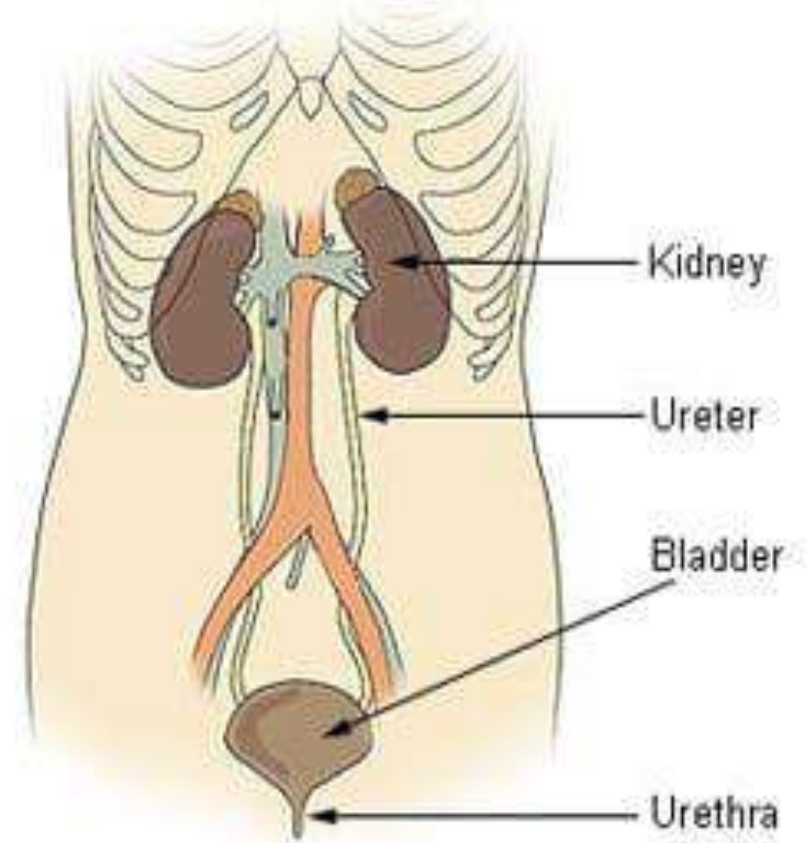
- Explain the different ways of urine collection.
- Explain the significance of the components of a urinalysis.
- List reference values for routine urinalysis.
- Explain at least factors that can influence the physical character of urine.

Introduction

The urinary system comprises:

- ❖ Two Kidneys.
- ❖ Ureters.
- ❖ Bladder.
- ❖ Urethra.

Components of the Urinary System



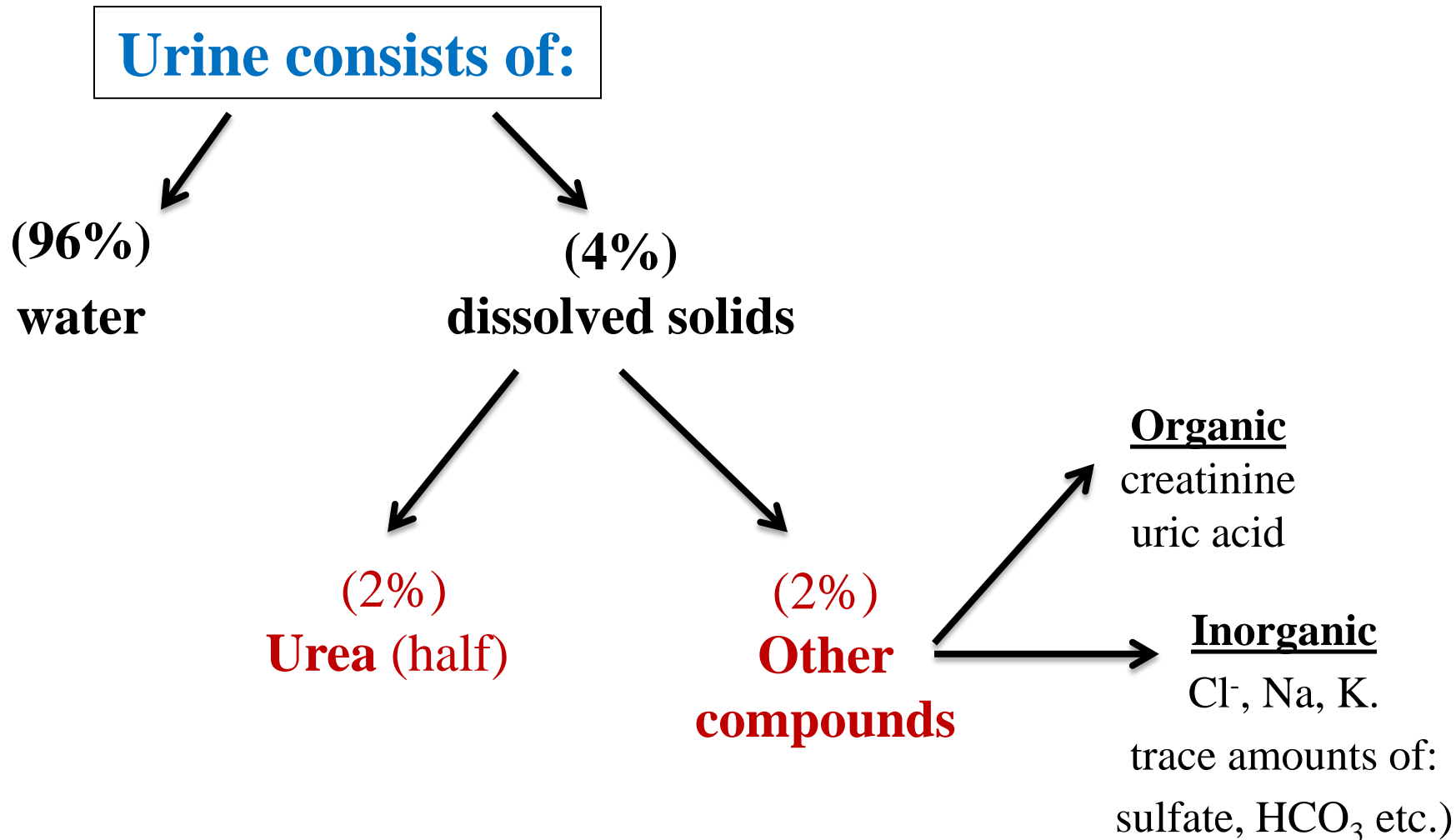
Functions of kidney

The kidneys filter unwanted waste materials from the blood and regulate the levels of water and chemicals in the body. About 99% of the fluid circulating through the kidneys is reabsorbed into the blood with the remaining excreted as urine.

Approximately 1000 liters of blood filtered through the kidneys produces one liter of urine.

Urine

- Urine carries waste products and excess water out of the body.



Factors affect on urine constituents:

- Dietary intake.
- Physical activity.
- Body metabolism.
- endocrine function.
- Others.

Urinalysis

Analysis defined as identification or separation of ingredients of a substance

A urinalysis requires (3) types of examination:

- **Physical examination:** To note color, odor, appearance and volume.
- **Chemical examination:** Tests include, pH, specific gravity protein, glucose, ketones, nitrite, etc...
- **Microscopic examination:** Sediment is examined for red blood cells, white blood cells, epithelial cells, casts, bacteria, yeast, and crystals.

Types of urine specimens

Over the course of a **24-hour** period, the composition and concentration of urine changes continuously. For this reason, various types of specimens may be collected. Generally, about **10 mL** of urine is required for routine urinalysis. Urine specimens should be refrigerated if they cannot be examined within **2 hours** **because** urine begins to break down after that time, becoming more alkaline, and rendering some urine tests inaccurate.

Urine collection

There are (3) ways to obtain a urine specimen:

1. Spontaneous voiding.
2. Urethral catheterization.
3. Supra pubic bladder puncture.

Physical examination of urine

- The first step in physical analysis is obtaining an appropriate specimen.

Reference values for normal urinalysis

Characteristic	Expected measurement
Color	Pale yellow to amber
Appearance	Clear to slightly hazy
Odor	Slight
Volume	1500 mL /24 hours (750-2500 range)
pH	Average 5-6
Specific gravity	Usually 1.015 with normal fluid intake

❑ Colour

- Colour of the urine varies from colourless to pale yellow or amber.

❑ Appearance

- Should be clear but may be slightly cloudy. Cloudy urine (white or yellow) may be evidence of infection with pus or microscopic blood present, but it can also be caused by kidney stones, foods, vaginal discharge, and dehydration.

❑ Odor

- Should be very slight, but some foods and medications, such as estrogen, may affect odor and metabolic disorders may produce a strong or distinctive urine odor.

❑ Volume

- Volume of urine for a healthy adult is about 750 and 2500 mL of urine in 24 hours, or approximately 25 to 30 mL per hour.

❑ Urine pH

- Urine pH measures the acidity of urine to determine if it is acidic or alkaline and serves as a screening test for renal, and metabolic disorders along with other tests.



Practical Physiology

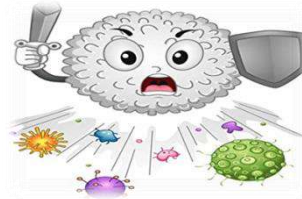
Lec. 9

White Blood Cells



قسم تقنيات التخدير
المرحلة الاولى
2020-2021

By- Ph.D. Shaymaa A. Ahmed

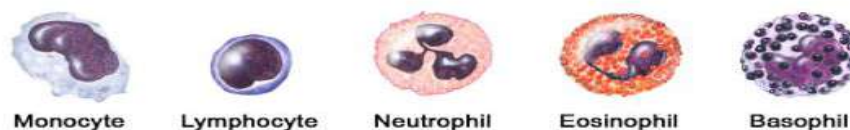


Introduction:

The real value of *White blood cells*, also called leukocytes, is that being the *mobile units* of the body's protective system against infectious agents such as bacteria, viruses, fungi, toxins and other foreign invaders that can sometimes cause lethal disease conditions.

Types of leukocytes:

The five main types of white blood cells are neutrophils, eosinophils, basophils, lymphocytes, and monocytes, (neutrophils, eosinophils, and basophils) are termed granulocytes (have granular cytoplasm) or polymorphonuclear leukocytes. The nuclei of most mature granulocytes are divided into two to five oval lobes connected by thin strands of chromatin. (Lymphocytes and monocytes) are referred to as agranular leukocytes, sometimes called mononuclear leukocytes do not have granules and have non lobular nuclei.



Normal values in blood:

The adult human being has about 4000-7000 WBCs / mm³ of blood. The normal percentages of the different types of WBCs are approximately the following:

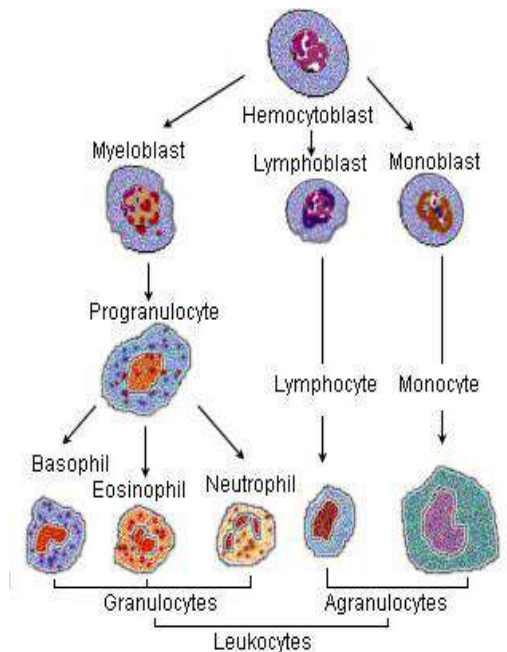
Neutrophils	62%
Eosinophils	2.3%
Basophils	0.4%
Monocytes	5.3%
Lymphocytes	30%

Sites of production:

The granulocytes and monocytes are formed only in the bone marrow. Lymphocytes are produced mainly in the lymph glands, spleen, thymus, tonsils, and various pockets of lymphoid tissue elsewhere in the body.

Leukopoiesis:

Leukopoiesis - the production, differentiation, and development of white blood cells. It begins with the division of hematopoietic stem cells (in bone marrow) to a committed stem cells that form the myeloblast, monoblast and the lymphoblast. Myeloblast differentiate by a series of divisions into promyelocyte then myelocyte to form (neutrophils, eosinophils, and basophils). Monoblast in turn, differentiate to promonocyte which form the (monocytes) while lymphoblast differentiates to prolymphocyte which form lymphocytes.



The WBCs formed in the bone marrow are stored within the marrow until they are needed in the circulatory system. Then, when the need arises, various factors cause them to be released. Such as colony stimulating factor (CSF) which is a substance released by WBCs and stimulates the bone marrow to increase white blood cell production as a response to an acute infection or inflammation.

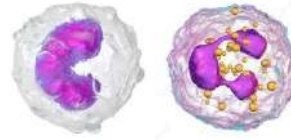
Life span:

The life of the **granulocytes** after being released from the bone marrow is normally 4 to 8 hours circulating in the blood and another 4 to 5 days in tissues where they are needed. In times of serious tissue infection, this total life span is often shortened to only a few hours. The **monocytes** also have a short transit time, 10 to 20 hours in the blood. Once in the tissues, they swell and become tissue macrophages; they can live for months unless

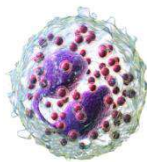
destroyed while performing phagocytic functions. The **lymphocytes** enter the circulatory system continually; they have life spans of weeks or months, depending on the body's need for these cells.

The defense mechanism in leukocytes:

Neutrophils and monocytes:



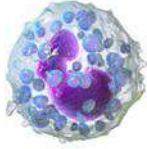
It is mainly the neutrophils and tissue macrophages (end-stage product of monocytes that enter the tissues from the blood.) that attack and destroy invading bacteria, viruses, and other foreign invaders by the **phagocytosis**, which means cellular ingestion of the foreign agent by engulfing and digesting it with digestive enzymes released from lysosomes (as proteolytic enzymes and lipase) and bactericidal agents released from peroxisomes (hydrogen peroxide H_2O_2 and hydroxyl ions OH^-) which are lethal to most bacteria, even in small quantities. A single neutrophil can usually phagocytize 3-20 bacteria before it becomes inactive and die. Monocytes are often capable of phagocytizing as many as 100 bacteria.



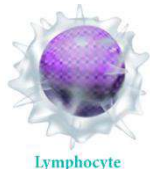
Eosinophils:

Eosinophils, are often produced in large numbers in people with parasitic infection, and they migrate into tissues diseased by parasites. Although most parasites are too large to be phagocytized by eosinophils or any other phagocytic cells, eosinophils attach themselves to the parasites and release substances that kill many of the parasites, substances such as hydrolytic enzymes released from their granules, or by releasing highly reactive forms of oxygen. Eosinophils also have a special propensity to collect in tissues in which allergic reactions occur, such as in the peribronchial tissues of the lungs in people with asthma and in the skin after allergic skin reactions.

Basophils:



Basophils liberate heparin into the blood. Heparin is a substance that can prevent blood coagulation. Basophils also release histamine, as well as smaller quantities of serotonin during inflammation of allergic reactions.



Lymphocytes:

Lymphocyte

Lymphocytes composed of B cells and T cells. The majority of lymphocytes are **T cells** they participate in cell-mediated immune defenses and are divided into several subtypes, such as helper T cells, cytotoxic T cells, and natural killer cells. Some lymphocytes are **B cells**, which are responsible for producing antibodies.

Good luck everyone

Ph.D. Shaymaa A. Ahmed





General Physiology

Lec. 10

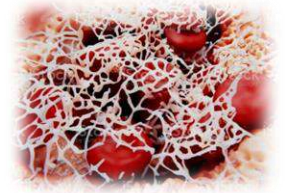
قسم تقنيات التخدير

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2020-2021



Hemostasis (Blood clotting mechanism)



By- Ph.D. Shaymaa A. Ahmed

Introduction:

The term *hemostasis* means prevention of blood loss or in other words (the cessation of blood loss from a damaged vessel) can be organized into four separate but interrelated events:

- ★ (1) Vascular constriction.
- ★ (2) Formation of a platelet plug.
- ★ (3) Formation of a blood clot as a result of blood coagulation.
- ★ (4) Breakdown of blood clot (fibrinolysis).

STEP 1. Vascular constriction

Immediately after tissue injury, blood flow through the disrupted vessel is slowed because of the contraction of the smooth muscles in vessels walls. The contraction is results from:

- ★ Local myogenic spasm of the blood vessels initiated by direct damage to the vascular wall.
- ★ Potent chemical substances released from damaged cells and platelet at the site of injured tissue that directly cause blood vessels to constrict. These include serotonin, thromboxane A₂, and epinephrine.
- ★ Nervous reflexes. The nervous reflexes are initiated by pain nerve impulses or other sensory impulses that originate from the traumatized vessel or nearby tissues.

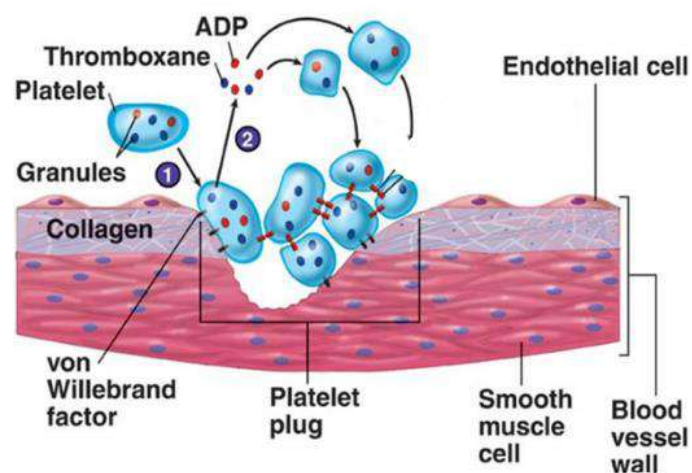


STEP 2• Formation of platelet plug

To understand this process, it is important that we first discuss the nature of platelets themselves:

Platelets (also called thrombocytes) are irregularly shaped, disk-like fragments of their precursor cell, the megakaryocyte (hematopoietic cells bone marrow). The normal concentration of platelets in the blood is between 150,000 and 300,000 per microliter. Thrombopoietin hormone which is mainly generated by the liver and the kidneys as a response to low numbers of circulating platelets, stimulate megakaryocytes to release platelets within the bone marrow. It has a life span in the blood of 8 to 12 days. Platelets have no defined nucleus but possess important proteins, which are stored in intracellular granules and secreted when platelets are activated during coagulation.

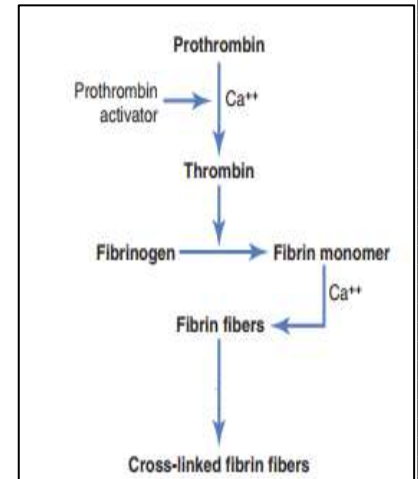
Platelet release granules that contain multiple active factors; they become sticky so that they adhere to collagen in the injured endothelial cells of the vessel wall. Endothelial cells and megakaryocytes released a critical substance called von Willebrand factor that enhances platelet adhesion to the endothelium by forming a bridge between platelet surface receptors and collagen. Platelets also secrete large quantities of ADP and thromboxane A₂ enzyme which activates the stickiness of additional platelets causes them to adhere to the original activated platelets thus forming a platelet plug.



STEP 3• Formation of blood clot

Clotting takes place in three essential steps:

- 🦋 Formation of a substance called **prothrombin activator** from platelet after they bind to the collagen fibers
- 🦋 The prothrombin activator catalyzes conversion of **prothrombin** into **thrombin**, in the presence of (Ca^{++}) stores in the mitochondria, endoplasmic reticulum, and Golgi apparatus of the platelets, prothrombin form in liver and require vitamin K.
- 🦋 The thrombin acts as an enzyme to convert **fibrinogen** into **fibrin fibers** that form a highly organized and firm network, which traps more platelets, red blood cells, and leukocytes at the site of vascular damage, thereby forming a stable blood clot.

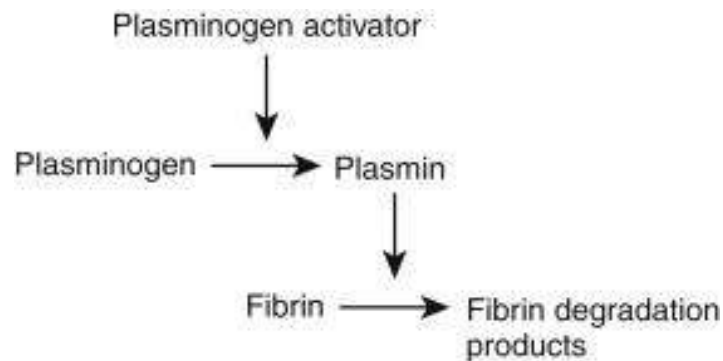


➡ This table shows clotting factors in blood and their synonyms:

Clotting Factor	Synonyms
Fibrinogen	Factor I
Prothrombin	Factor II
Tissue factor	Factor III; tissue thromboplastin
Calcium	Factor IV
Factor V	Proaccelerin; labile factor; Ac-globulin (Ac-G)
Factor VII	Serum prothrombin conversion accelerator (SPCA); proconvertin; stable factor
Factor VIII	Antihemophilic factor (AHF); antihemophilic globulin (AHG); antihemophilic factor A
Factor IX	Plasma thromboplastin component (PTC); Christmas factor; antihemophilic factor B
Factor X	Stuart factor; Stuart-Prower factor
Factor XI	Plasma thromboplastin antecedent (PTA); antihemophilic factor C
Factor XII	Hageman factor
Factor XIII	Fibrin-stabilizing factor
Prekallikrein	Fletcher factor
High-molecular-weight kininogen	Fitzgerald factor; HMWK (high-molecular-weight kininogen)
Platelets	

STEP 4. Fibrinolysis

Fibrinolysis is the process of breaking down the fibrin clot. The main enzyme in fibrinolysis is **plasmin**, which cuts the fibrin mesh at various places, leading to the formation of circulating fragments that are cleared by other proteases or by the kidney and liver. Plasmin circulates as inactive proenzyme **plasminogen**, which converted to plasmin by a **tissue plasminogen activator (TPA)**, that released by activated endothelial cells.



Thank you

الأزهارُ تحتاجُ وقتًا لتُزهر؛
*Flowers need time
to bloom.*
وكذلك **أنتم**
so do you.

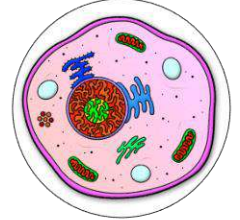
د. شيماء

General Physiology

By- Ph.D. Shaima'a Al-jubouri

Lec. 1

CELL PHYSIOLOGY



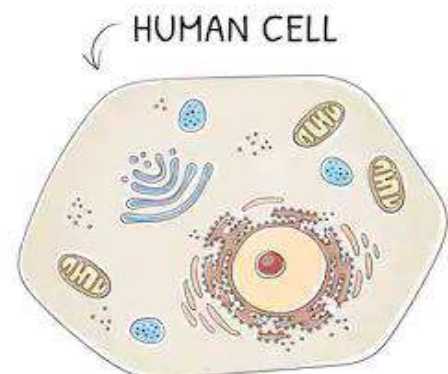
PHYSIOLOGY:

Physiology is the science that seeks to explain the physical and chemical mechanisms that are responsible for the origin, development, and progression of life.

Each type of life, from the simplest virus to the complicated human being, has its own functional characteristics. Therefore, the vast field of physiology can be divided into viral physiology, bacterial physiology, cellular physiology, plant physiology, invertebrate physiology, vertebrate physiology, mammalian physiology, human physiology, and many more subdivisions.

THE CELL:

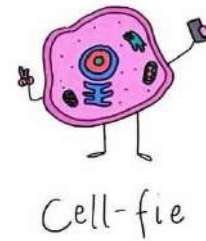
The basic living unit of the body is the cell (the structural and functional unit of life). Each organ is an aggregate of many different cells, each type of cell is specially adapted to perform various functions such as metabolism, transportation and secretion of substances.



Cells main functions:

It provide six functions:

- ✿ Structure and support
- ✿ Facilitate growth through mitosis
- ✿ Allow passive and active transport
- ✿ Produce energy
- ✿ Create metabolic reactions
- ✿ Aid in reproduction.

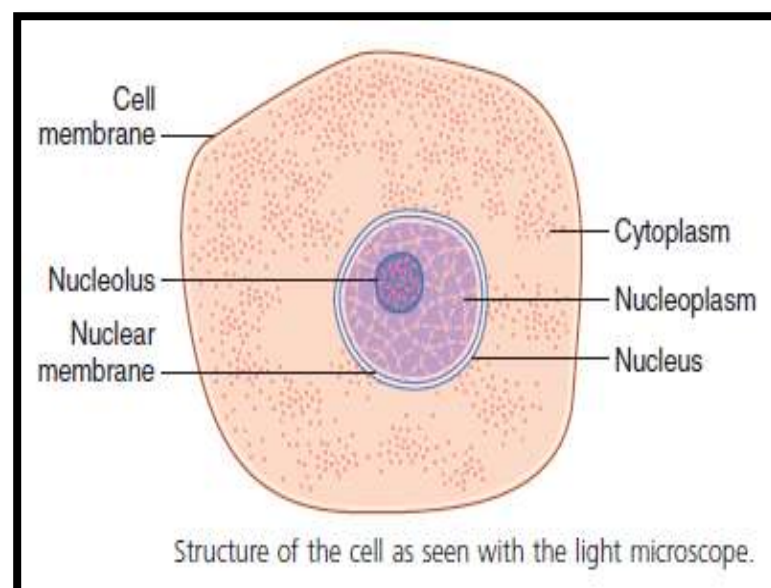


Organization of the Cell:

A typical cell formed by two major parts: the *nucleus* and the *cytoplasm*. The nucleus is separated from the cytoplasm by a *nuclear membrane*, and the cytoplasm is separated from the surrounding fluids by a *cell membrane*. The different substances that make up the cell are collectively called *protoplasm*.

Protoplasm is composed mainly of five basic substances:

- ✿ Water
- ✿ Electrolytes
- ✿ Proteins
- ✿ Lipids
- ✿ Carbohydrates

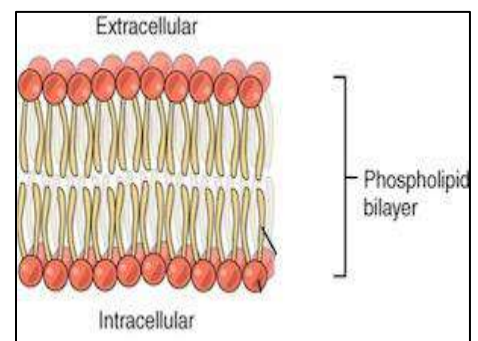


CELL MEMBRANE:

The cell membrane (also called the *plasma membrane*) which envelops the cell, is a thin, pliable, elastic structure. It is composed almost entirely of proteins and lipids. Membranes cover most organelles of the cell, which composed of the same structural components. The plasma membrane consists of:

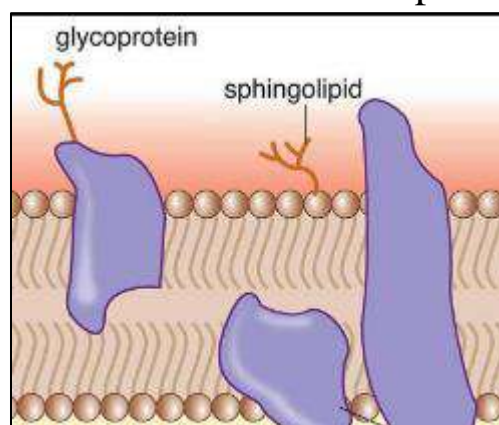
- **Cell membrane lipids:** The basic structure of cell membrane is a **lipid bilayer**, which is a thin, double-layered film of lipids—each layer only one molecule thick—that is continuous over the entire cell surface. The basic lipid bilayer is composed of three main types of lipids: **phospholipids**, **sphingolipids**, and **cholesterol**.

Phospholipids are the most abundant of the cell membrane lipids. The lipid layer in the middle of the membrane is impermeable to the usual water-soluble substances, such as ions, glucose, and urea. Conversely, fat-soluble substances, such as oxygen, carbon dioxide, and alcohol, can penetrate this portion of the membrane with ease.



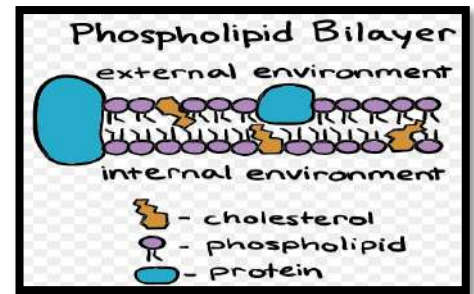
Sphingolipids, present in small amounts in the cell membranes, especially nerve cells. It have several functions, including

- ✿ Protection from harmful environmental factors
- ✿ Signal transmission
- ✿ As adhesion sites for extracellular proteins.



Cholesterol molecules in the membrane are also lipid in nature because their steroid nuclei are highly fat-soluble.

- ✿ They mainly help determine the degree of permeability (or impermeability) of the bilayer to water-soluble constituents of body fluids.
- ✿ Cholesterol controls much of the fluidity of the membrane as well.

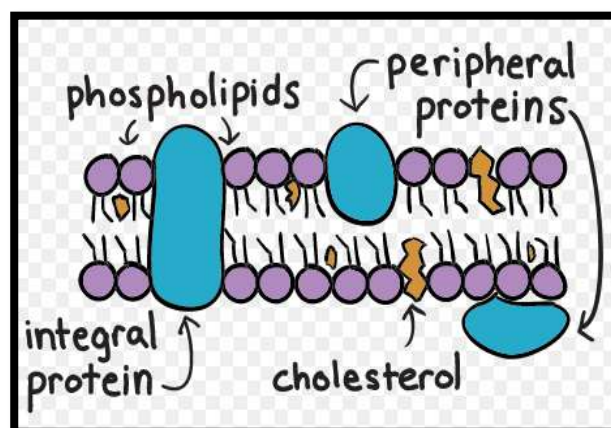


● **Cell Membrane Proteins:** There are two types of cell membrane proteins:

Integral proteins: that protrude all the way through the membrane. It has several functions such as:

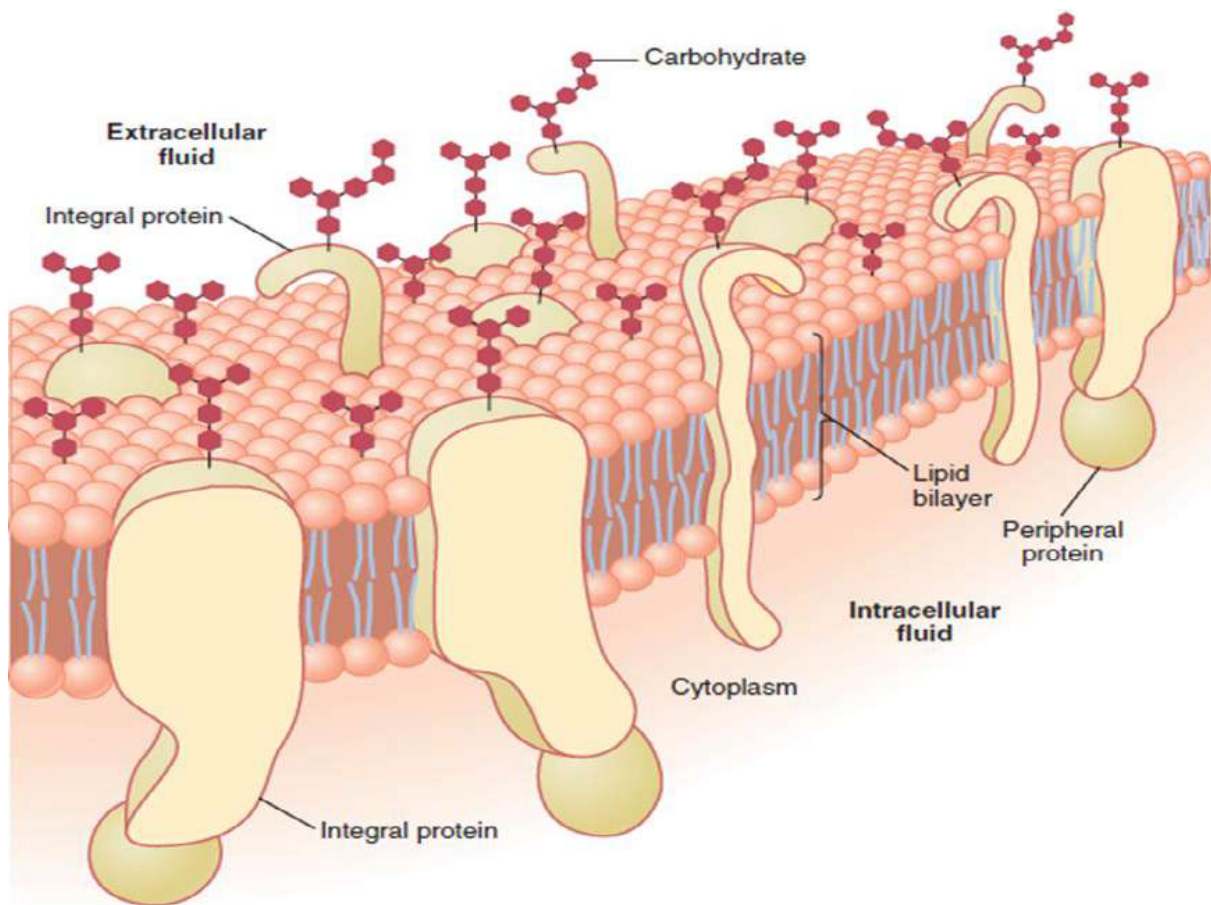
- ✿ Provide structural *channels* (or *pores*) through which water molecules and water-soluble substances, especially ions, can diffuse between the extracellular and intracellular fluids.
- ✿ Act as carrier proteins for transporting substances that otherwise could not penetrate the lipid layer.
- ✿ Serve as receptors for water-soluble chemicals, such as peptide hormones, that do not easily penetrate the cell membrane.

Peripheral protein molecules: are often attached to the integral proteins. These peripheral proteins function almost entirely as enzymes or as controllers of transport of substances through the cell membrane “pores.”



● **Cell membrane Carbohydrates:** Membrane carbohydrates occur almost invariably in combination with proteins or lipids in the form of *glycoproteins* or *glycolipids*. The entire outside surface of the cell often has a loose carbohydrate coat called the **glycocalyx**. The carbohydrate moieties attached to the outer surface of the cell have several important functions:

- ✿ Many of them have a negative electrical charge, which gives most cells an overall negative surface charge that repels other negative objects.
- ✿ The glycocalyx of some cells attaches to the glycocalyx of other cells, thus attaching cells to one another.
- ✿ Many of the carbohydrates act as *receptor substances* for binding hormones, such as insulin.
- ✿ Some carbohydrate moieties enter into immune reactions.

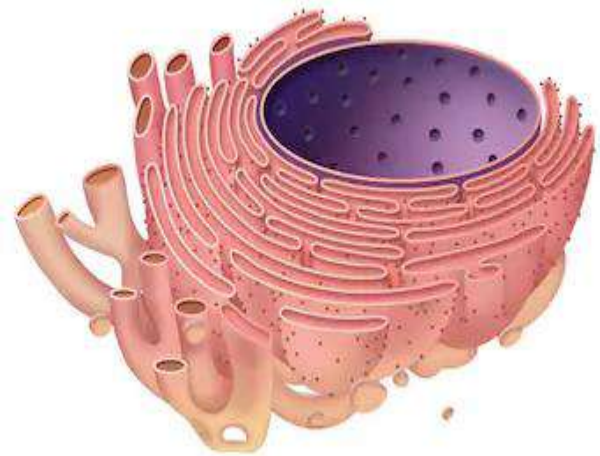
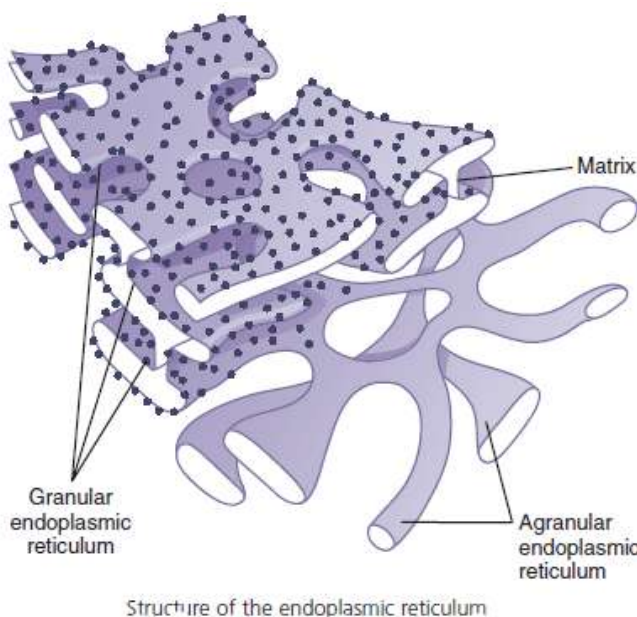


Structure of the cell membrane

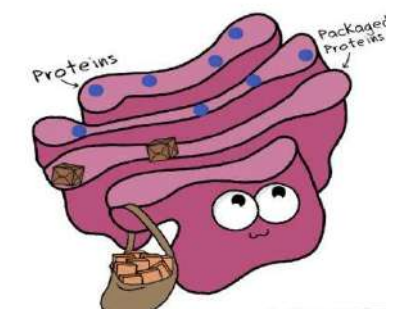
➡ CYTOPLASM AND ITS ORGANELLES:

The cytoplasm is filled with both minute and large dispersed particles and organelles. The clear fluid portion of the cytoplasm in which the particles are dispersed is called cytosol. It includes:

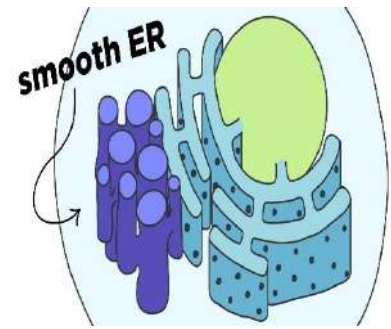
- **Endoplasmic Reticulum:** network of tubular and flat vesicular structures in the cytoplasm. This organelle helps process molecules made by the cell and transports them to their specific destinations inside or outside the cell.



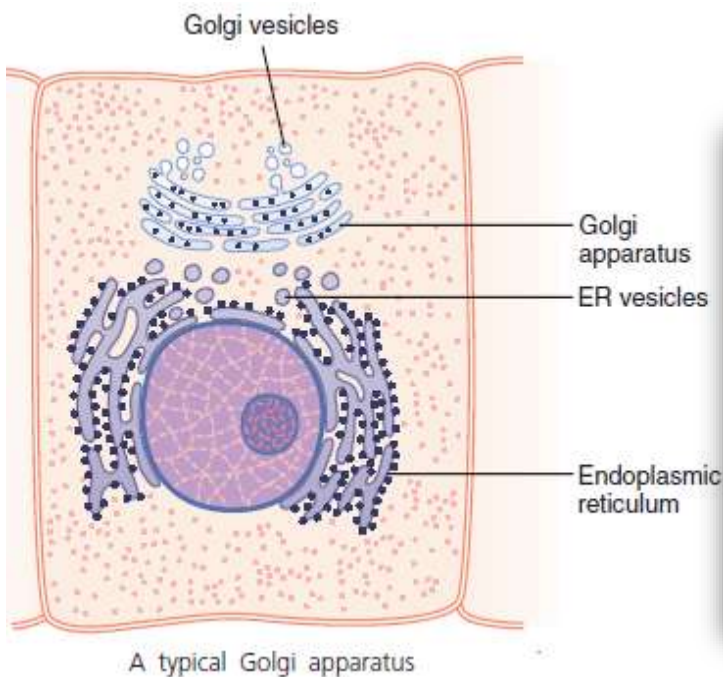
Granular Endoplasmic Reticulum: Attached to the outer surfaces of many parts of the endoplasmic reticulum, large numbers of minute granular particles that lack membranes called **ribosomes**. Where these are present, the reticulum is called the *granular endoplasmic Reticulum*. Its function is to synthesize new protein molecules.



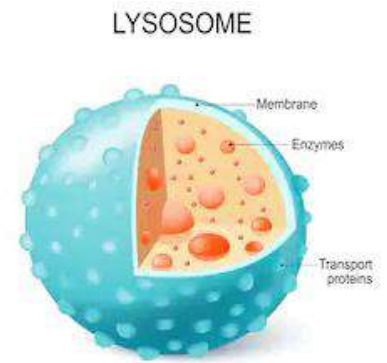
A granular Endoplasmic Reticulum: Part of the endoplasmic reticulum has no attached ribosomes. This part is called the *agranular*, or *smooth endoplasmic reticulum*. its functions is synthesis of lipid substances.



- **Golgi apparatus:** Golgi apparatus is usually composed of four or more stacked layers of thin, flat, enclosed vesicles lying near one side of the nucleus. This apparatus is prominent in secretory cells, where it is located on the side of the cell from which the secretory substances are extruded. The Golgi apparatus functions in association with the endoplasmic reticulum; small “transport vesicles” (endoplasmic reticulum vesicles) continually pinch off from the endoplasmic reticulum then fused with the Golgi apparatus. So, substances entrapped in the ER vesicles are transported to the Golgi apparatus. The transported substances are then processed in the Golgi apparatus to form lysosomes, secretory vesicles and other cytoplasmic components



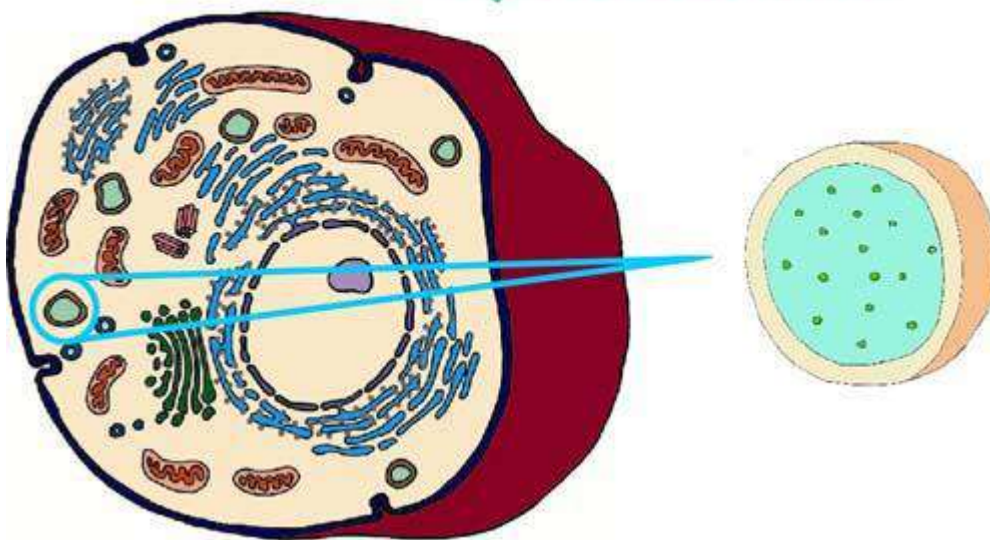
● **Lysosomes:** Its vesicular organelles that form by breaking off from the Golgi apparatus and then dispersing throughout the cytoplasm. The lysosomes provide an *intracellular digestive system* that allows the cell to digest:



- ✿ Damaged cellular structures.
- ✿ Food particles that have been ingested by the cell
- ✿ Unwanted matter such as bacteria.

It is surrounded by a typical lipid bilayer membrane and is filled with large numbers of small granules, which are protein aggregates of as many as 40 different hydrolase (digestive) enzymes. A hydrolytic enzyme is capable of splitting an organic compound into two or more parts (protein, glycogen, and lipids). The membrane surrounding the lysosome prevents the hydrolytic enzymes from coming in contact with other substances in the cell and therefore prevents their digestive actions.

Lysosome



● Mitochondria:

It also called the “power houses” of the cell. Without them, cells would be unable to extract enough energy from the nutrients. Mitochondria total varies from less than a hundred up to several thousand. They are also variable in size and shape. Some are globular in shape, Whereas others are elongated still others are branching and filamentous.



The basic structure of the mitochondrion is composed mainly of two lipid bilayer– protein membranes: an *outer membrane* and an *inner membrane*. Many infoldings of the inner membrane form shelves or tubules called *cristae* onto which oxidative enzymes are attached.

In addition, the inner cavity of the mitochondrion is filled with a *matrix* that contains large quantities of dissolved enzymes that are necessary for extracting energy from nutrients. These enzymes operate in association with the oxidative enzymes on the cristae to cause oxidation of the nutrients, thereby forming carbon dioxide and water and at the same time releasing energy.

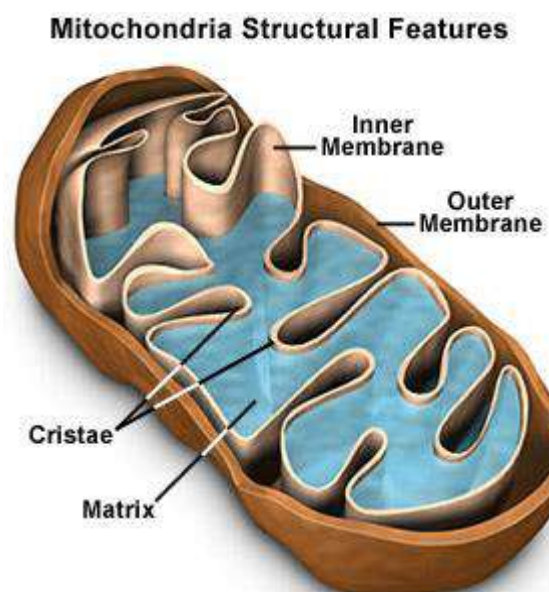
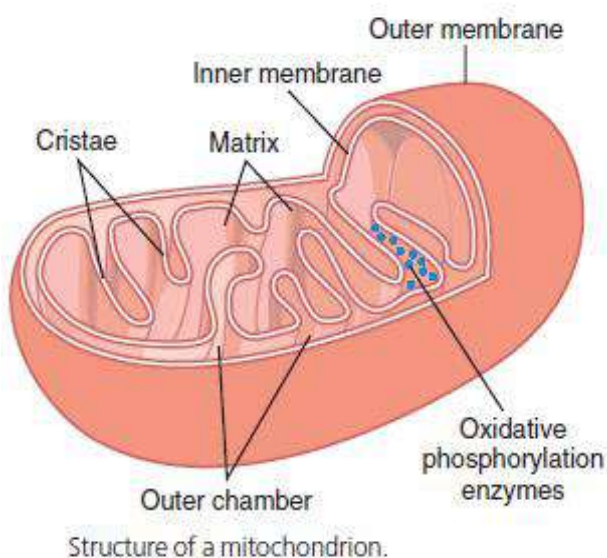
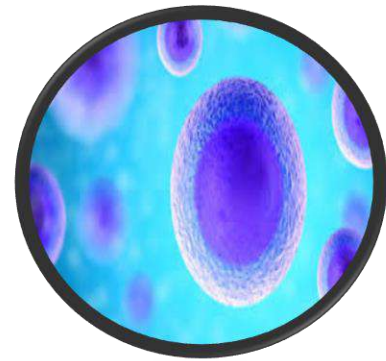
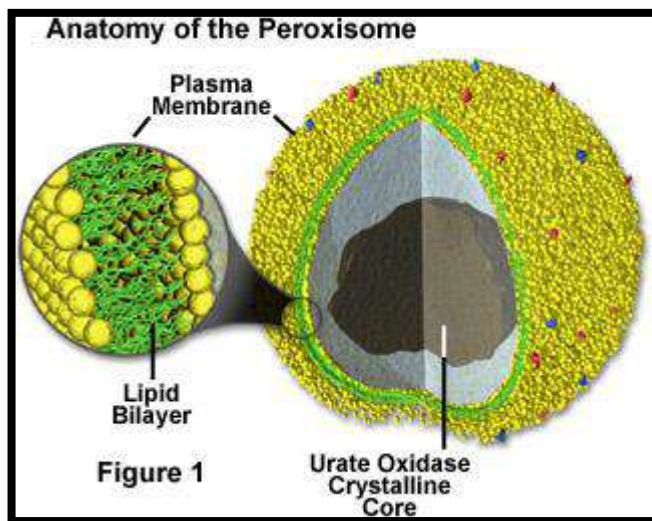


Figure 1

● Peroxisomes:

Peroxisomes are similar physically to lysosomes, but they are different in two important ways. **First**, they are believed to be formed by self-replication (or perhaps by budding off from the smooth endoplasmic reticulum) rather than from the Golgi apparatus. **Second**, they contain oxidases rather than hydrolases. Several of the oxidases are capable of oxidize various organic substances producing H_2O_2 of combining oxygen with hydrogen ions derived from different intracellular chemicals to form hydrogen peroxide(H_2O_2) which is highly oxidizing substance and is used to oxidize many substances that might otherwise be poisonous to the cell.



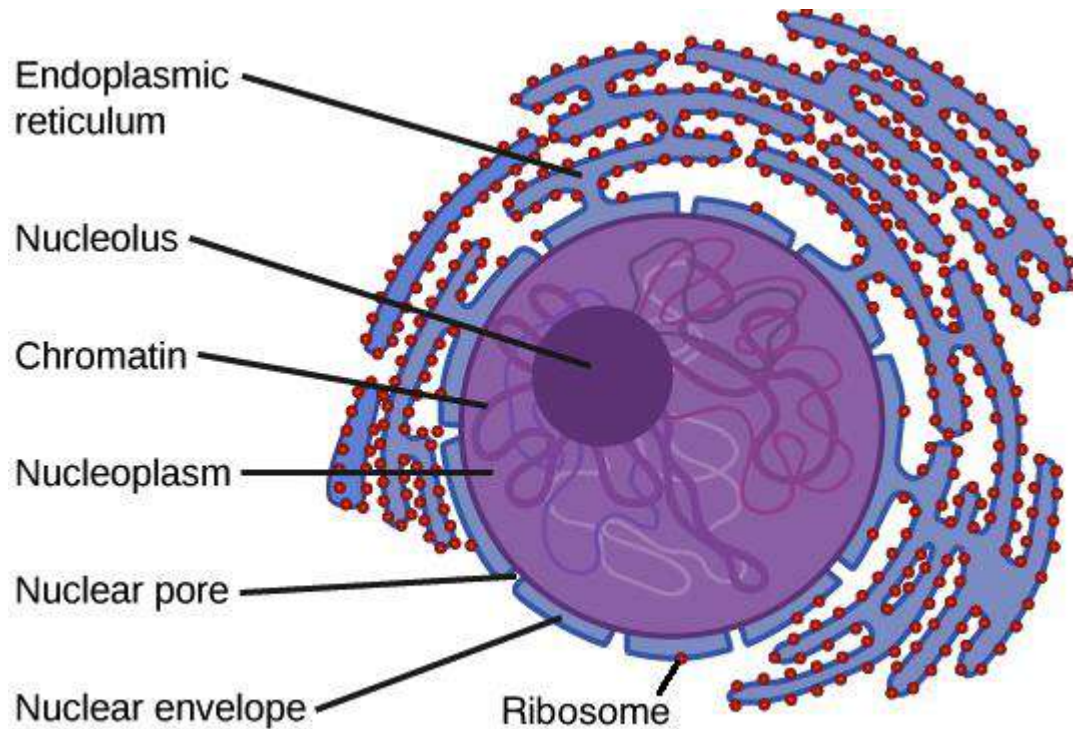
● Nucleus:

The nucleus, which is the control center of the cell, sends messages to the cell to grow and mature, to replicate, or to die. Briefly, the nucleus contains large quantities of DNA, which comprise the *genes*. The genes determine the characteristics of the cell's proteins, including the structural proteins, as well as the intracellular enzymes that control cytoplasmic and nuclear activities.



Nucleus surrounded by a **Nuclear Membrane** also called the nuclear envelope; a bilayer membrane that is continuous with the endoplasmic reticulum of the cell cytoplasm, it is penetrated by several thousand of nuclear pores.

The nuclei of most cells contain one or more highly staining structures called nucleoli. Some RNA is stored in the nucleoli.



GOOD LUCK

Ph.D. Shaima'a Al-jubouri





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General Physiology

Lec. 1

Movement of fluids and substances across the cell membrane

By- Ph.D Shaima'a A. Aljubouri

Introduction:

All cells must import oxygen, sugars, amino acids, and small ions and export carbon dioxide, metabolic wastes, and secretions. At the same time, specialized cells require mechanisms to transport molecules such as enzymes, hormones, and neurotransmitters. The movement of large molecules is carried out by endocytosis and exocytosis, that is, the transfer of substances into or out of the cell, respectively, by vesicle formation and vesicle fusion with the plasma membrane. Cells also have mechanisms for the rapid movement of ions and solute molecules across the plasma membrane. These mechanisms are of two general types:

passive transport, which requires no direct expenditure of metabolic energy e.g. (diffusion and osmosis)

active transport, which uses metabolic energy to move solutes across the plasma membrane, e.g. (phagocytosis, endocytosis and exocytosis)

Passive transport: which includes the following mechanisms of movements:

Diffusion:

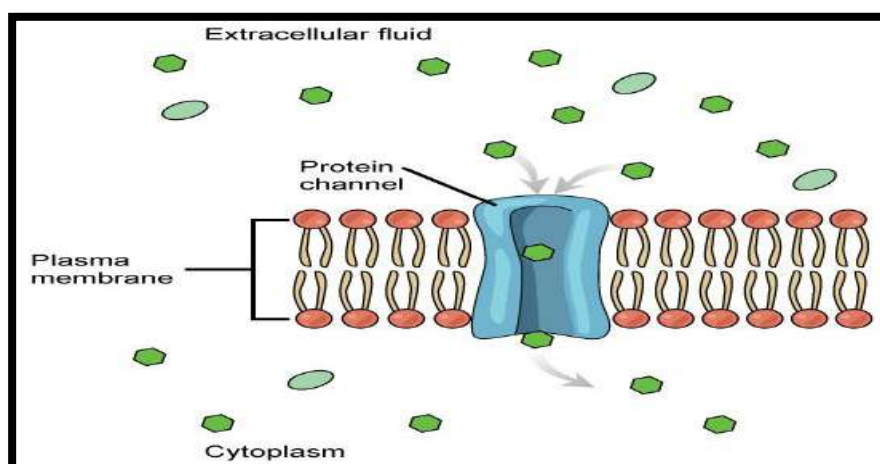
Is a continual random movement of molecules among one another in liquids or in gases from their area of higher concentration to their area of lower concentration. Diffusion through the cell membrane is divided into two subtypes, called *simple diffusion* and *facilitated diffusion*.

★ **Simple diffusion:**

Means that kinetic movement of molecules or ions occurs through a membrane opening or through intermolecular spaces without any interaction with carrier proteins in the membrane.

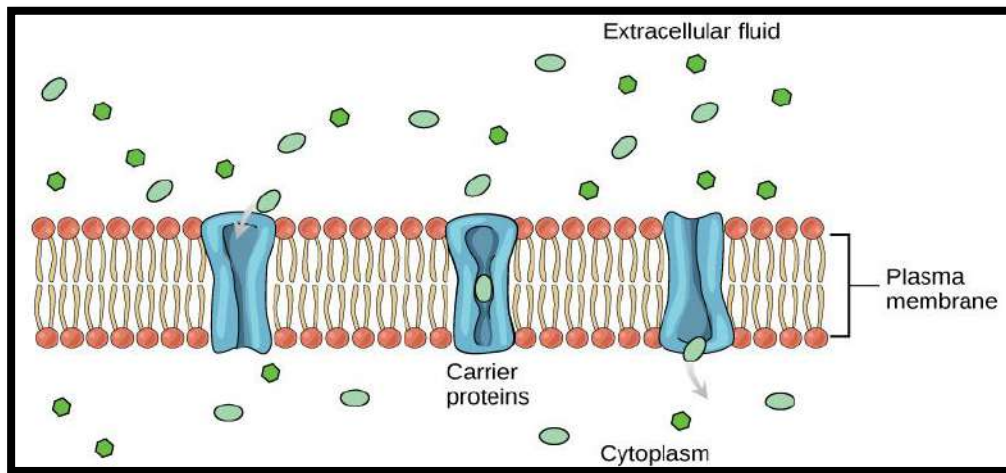
Movement occur all directions (randomly) and along the gradient through:

- ✓ The interstices of the lipid bilayer **or**
- ✓ The watery channels that penetrate all the way through some of the large transport proteins.



★ Facilitated diffusion

Facilitated diffusion is also called *carrier-mediated diffusion* because a substance transported in this manner diffuses through the membrane with the help of a specific carrier protein. That is, the carrier *facilitates* diffusion of the substance to the other side.



This type of diffusion is like other types of diffusion except for the carrier's ability to help the molecule move across the membrane. (Its move from higher to lower concentration). Due to molecule too large to pass through the pores and or not sufficiently soluble in the membrane lipids to diffuse through them. Among the many substances that cross cell membranes by facilitated diffusion are *glucose* and most of the *amino acids*.

➤➤ Diffusion of lipid-soluble substances through the lipid bilayer:

The lipid solubilities' of oxygen, nitrogen, carbon dioxide, and alcohols are high, and all these substances can dissolve directly in the lipid bilayer and diffuse through the cell membrane in the same manner that diffusion of water solutes occurs in a watery solution. The rate of diffusion of each of these substances through the membrane is directly proportional to its lipid solubility.

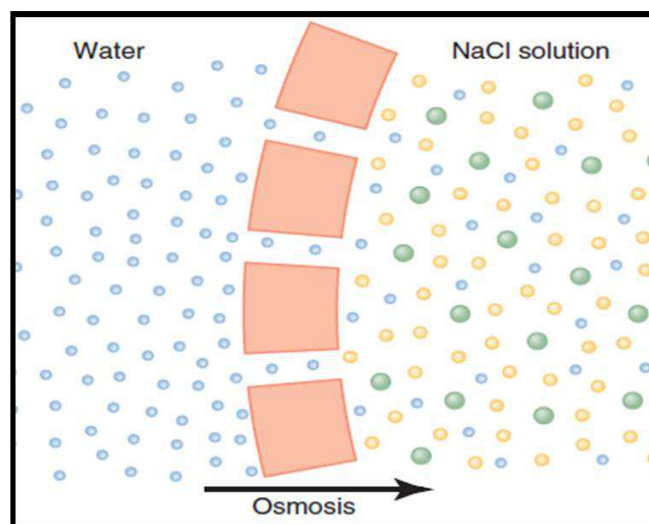
➤➤ Diffusion of water and other lipid-insoluble molecules through protein channels:

Many of the body's cell membranes contain protein "pores" called *aquaporins* that selectively permit rapid passage of water through the membrane. Other lipid-insoluble molecules can pass through the protein pore channels in the same way as water molecules if they are water soluble and small enough.

Osmosis:

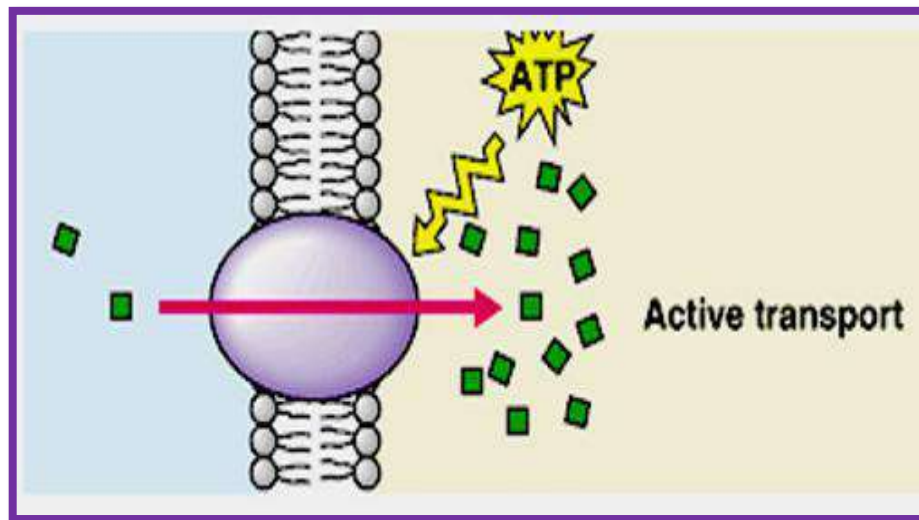
The spontaneous movement of water from a solution of high water concentration (low concentration of solute) toward a solution with a lower concentration of water (high solute concentration). Specific membrane proteins that function as water channels explain the rapid movement of water across the plasma membrane. These water channels are small, integral membrane proteins known as **aquaporins**.

Osmosis is a passive transport mechanism that tends to equalize the total solute concentrations of the solutions on both sides of every membrane.



Active transport:

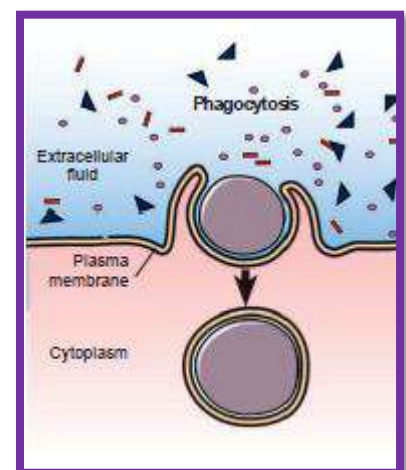
Means movement of ions or other substances across the membrane in combination with a carrier protein from a low-concentration state to a high-concentration state. This movement requires an additional source of energy that is usually derived from a substance called adenosine triphosphate, or ATP.



Different substances that are actively transported through at least some cell membranes include sodium, potassium, calcium, iron, hydrogen, chloride, iodide, and urate ions, several different sugars, and most of the amino acids.

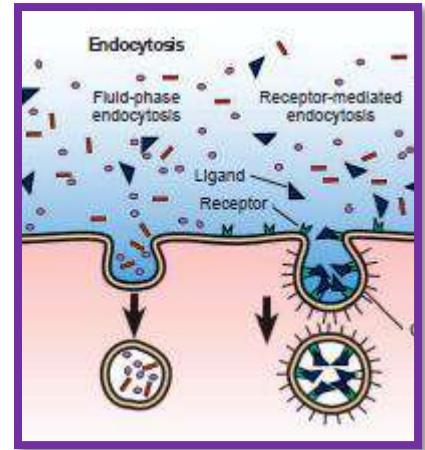
Phagocytosis:

It is the ingestion of large particles or microorganisms, usually occurring only in specialized cells such as macrophages, which occurs only after the extracellular particle has bound to the extracellular surface. The particle is then enveloped by expansion of the cell membrane around it.



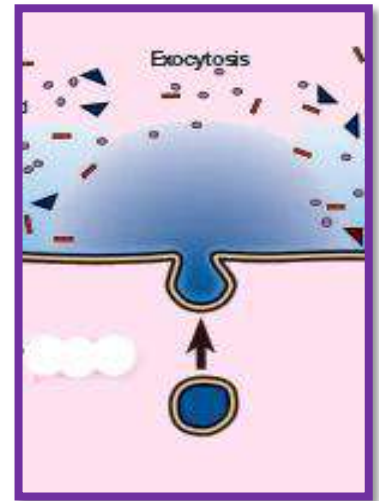
Endocytosis:

The process in which a region of the plasma membrane is pinched off to form an endocytic vesicle inside the cell. During vesicle formation, some fluid, dissolved solutes, and particulate material from the extracellular medium are trapped inside the vesicle and internalized by the cell.



Exocytosis:

It is the reverse of endocytosis. The formation of important macromolecules that are destined for export from the cell. These molecules are packed inside transport vesicles and move to the cell surface, fuse with the cell membrane, and release their contents outside the cell.



Good Luck!

د. شيماء



General Physiology

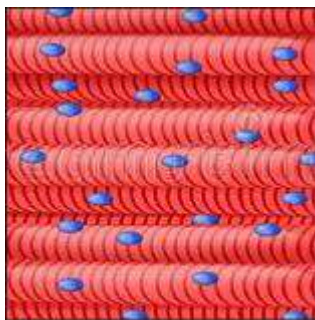
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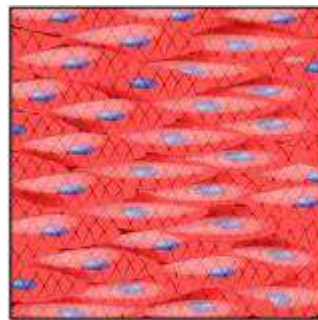
Muscular tissues:

Types and Characteristic

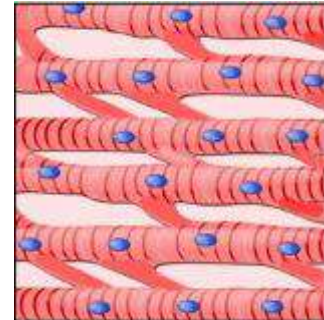
By- Ph.D Shaima'a A. Aljubouri



Skeletal muscle



Smooth muscle



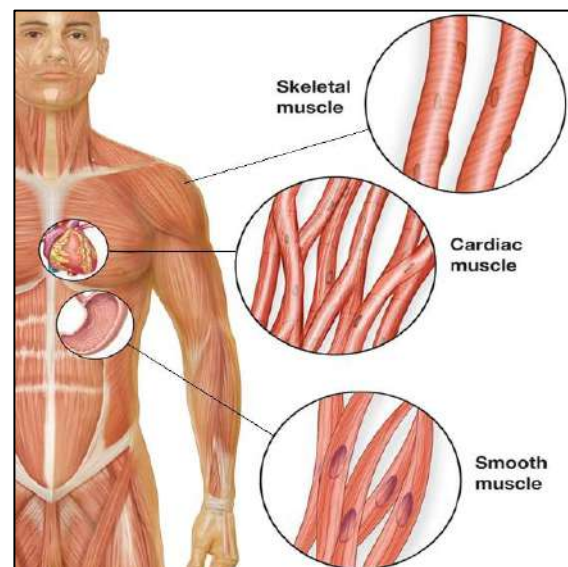
Cardiac muscle

Introduction:

Muscles are organs composed of cells that possess the ability to contract. Therefore, it convert the chemical energy to mechanical energy, generating force and movement.

Muscle is commonly classified into subtypes based on anatomical location and appearance:

- ✿ Skeletal muscle
- ✿ Cardiac muscle
- ✿ Smooth muscle

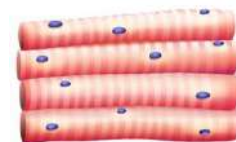


➡ Main functions of muscular system:

- ✿ Mobility and posture: muscular tissues allow movement and the skeletal muscles keep the body in the correct position (sitting or standing).
- ✿ Circulation: Heart muscle circulates blood, smooth muscle (vessels) plays a role in maintains blood pressure and prevents blood loss or dehydration
- ✿ Respiration: use diaphragm, intercostal muscles
- ✿ Digestion: Smooth muscles (movement, digestion of foods).
- ✿ Urination: Smooth muscles and skeletal muscles and nerves work together to hold and release urine from the bladder.
- ✿ Childbirth: Smooth muscles (uterus) expand and contract to push the baby during childbirth.
- ✿ Vision: Six skeletal muscles around the eye control its movements
- ✿ Organ protections: Muscles protect the bones and organs by absorbing shock and reducing friction in joints.
- ✿ Temperature regulation: 85% of the heat generates in body comes from contracting muscles.



SKELETAL MUSCLE



Skeletal muscle

About 40 percent of the body is skeletal muscle that is primarily associated with the bones of the skeleton.

Skeletal muscle is a striated, voluntary (neurogenic) muscle needs nerve supply to work, composed of numerous muscle cells, also called **muscle fibers**, which consist of a single cell,

multinucleated, cylindrical shape surrounded by cell membrane called **sarcolemma**, the nuclei occupy positions near the periphery of the fiber, each fiber is usually innervated by only one nerve ending.

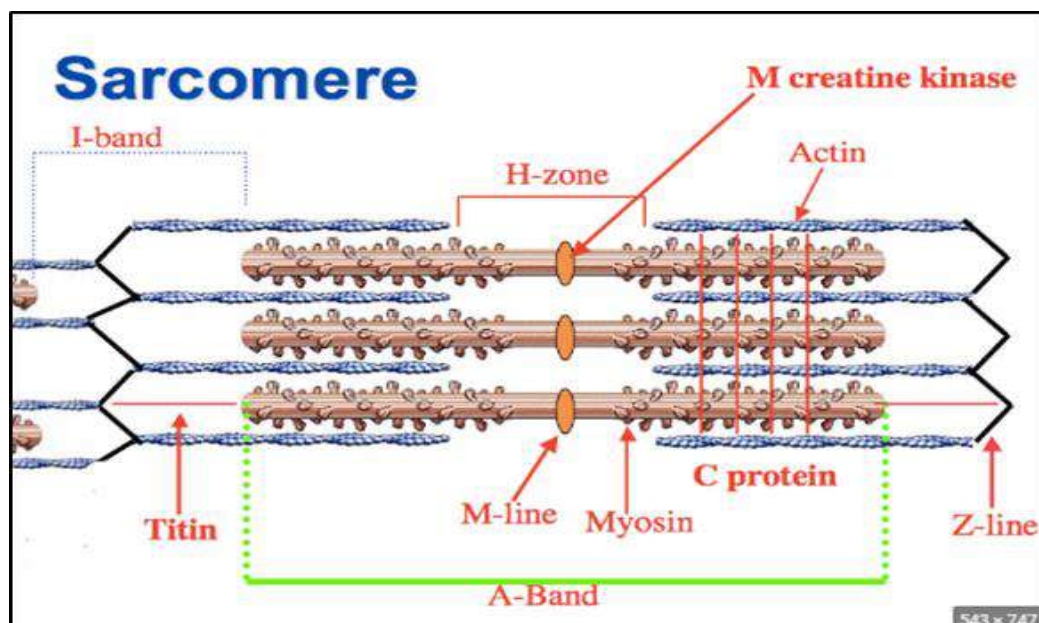
Each muscle fibers is made up of smaller subunits called (myofibrils), each myofibrils composed of contractile protein. Skeletal muscle has an abundant supply of mitochondria, which are vital for supplying chemical energy in the form of adenosine triphosphate (ATP) to the contractile system.

Actin and myosin filaments:

Myofibril is composed of **myosin filaments (thick filaments)** and **actin filaments (thin filaments)**, which are large polymerized protein molecules that are responsible for muscle contraction.

Each myofibril has alternating light and dark bands, the dark band of a myofibril is called an **A bands** (contain the myosin filaments) which divided at its center by a narrow, lighter-colored region called an **H zone**.

In many skeletal muscles, a prominent **M line** is found at the center of the H zone. Low-density **I bands** (contain the actin filaments) lie between the A bands. Crossing the center of I band is a dark structure called a **Z line** (or *Z disk*).

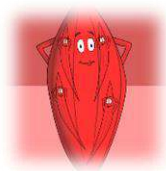


Sarcomere:

The fundamental repeating unit of the myofibril bands is called a **sarcomere** and is defined as the space between (and including) two successive Z lines.

Sarcolemma:

The sarcolemma is a thin membrane enclosing a skeletal muscle Fiber, consists of plasma membrane, and an outer coat made up of a thin layer of polysaccharide material



SMOOTH MUSCLE:



Fusiform non-striated involuntary cells differ from skeletal and cardiac muscle fiber being much smaller. The contractile proteins are actin, myosin, and tropomyosin.

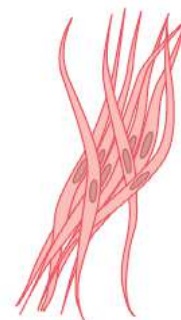
There are two main types of smooth muscle: the visceral (unitary) and the multiunit.

Multi - Unit Smooth Muscle.

Important characteristics of multi-unit smooth muscle fibers are: Composed of separate smooth muscle fibers, each fiber can contract independently of the others, and their control exerted mainly by nerve signals

The outer surfaces of smooth muscles fibers are covered by a mixture of fine collagen and glycoprotein that helps insulate the separate fibers from one another.

Some examples: the ciliary and iris muscle of the eye.



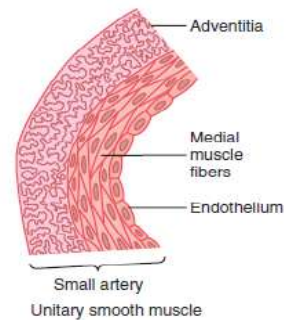
Multi-unit smooth muscle

Unitary Smooth Muscles:

Also called syncytial smooth muscle or visceral smooth muscle. Important characteristics are:

The smooth muscle fibers contract together as a single unit.

The fibers usually are arranged in sheets or bundles, and their cell membranes are adherent to one another at multiple points so that force generated in one muscle fiber can be transmitted to the next. In addition, the cell membranes are joined by many **gap junctions** through which ions can flow freely from one muscle cell to the next and cause the muscle fibers to contract together.



CARDIAC MUSCLE:

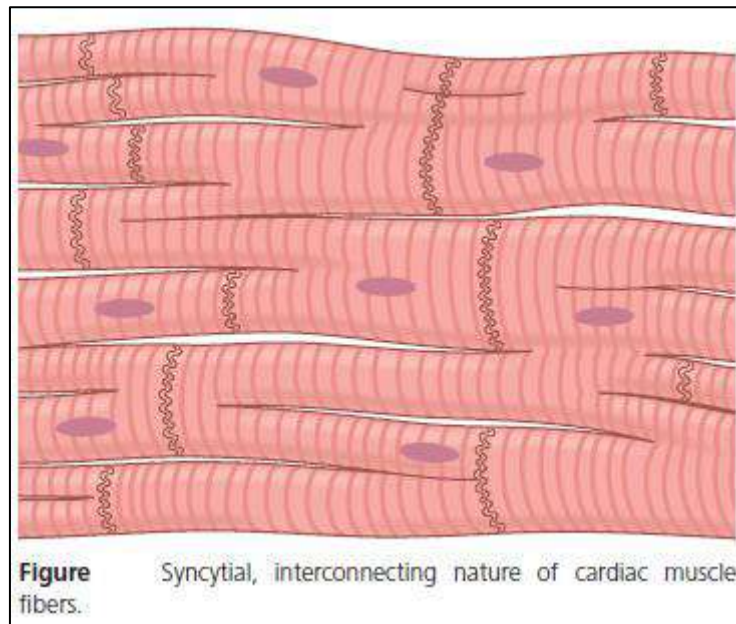


Cardiac muscle which is found exclusively in heart is branching, involuntary have single nucleus striated in the same manner as in skeletal muscle. Further, it has typical myofibrils that contain actin and myosin filaments almost identical to those found in skeletal muscle.

Cardiac muscle are connected by intercalated discs (they are actually cell membranes that separate individual cardiac muscle cells from one another). At each intercalated disc the cell membranes fuse with one another to form permeable “communicating” junctions (**gap junctions**) that allow rapid diffusion of ions.

Cardiac muscle act as a **single unit(syncytium)**, in which the cardiac cells are so interconnected that when one of these cells becomes excited, the action potential spreads to all of them,

The heart actually is composed of two syncytiums: the **atrial syncytium**, which constitutes the walls of the two atria, and the **ventricular syncytium**, which constitutes the walls of the two ventricles.



GOOD LUCK

Ph.D Shaima'a A. Aljubouri

Lec. 4

Muscles contraction



By- Ph.D. Shaimaa A. Aljubouri

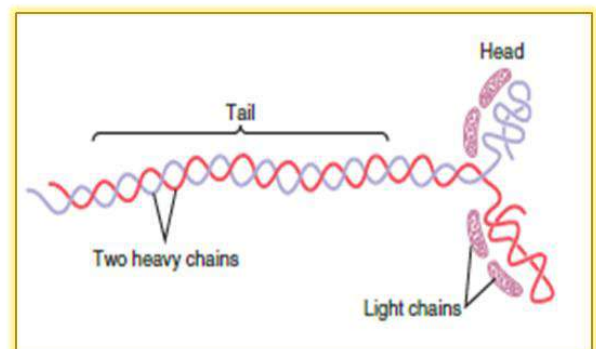
Introduction:

To understand the mechanism by which muscle contracts, it is first important to understand its structure. The striated muscles in our body are made up of many fibers. Inside these fibers are smaller units called myofibrils which are made of parallel thin and thick filaments. These filaments are arranged longitudinally in small units known as sarcomeres. These repeating sarcomeres give the muscle a striated appearance under microscopy, which accounts for its name

Molecular characteristics of the contractile filaments:



The thick filament: composed of several hundreds of **myosin** molecules, each molecule consists of two heavy polypeptide chains which is wrap spirally around each other to form the **tail**. At the end of each tail a globular polypeptide structure called **head**. Thus, there are two free heads at one end of the double-helix myosin molecule. The head of myosin molecule form the cross-bridges, which bind with actin.





The thin filament: composed of three proteins:



Actin: The backbone of actin filament is a double-stranded F-actin protein molecule. The two strands wound in a helix in the same manner as the myosin molecule. Actin contain active sites on its surface in which the cross –bridges of myosin attached



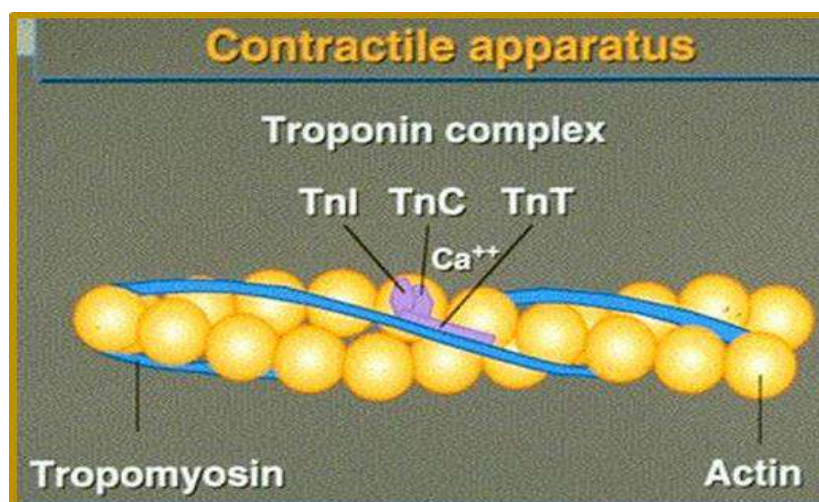
Tropomyosin: In the resting state, the tropomyosin molecules lie on top of the active sites of the actin strands so that attraction cannot occur between the actin and myosin filaments to cause contraction.



Troponin: These protein molecules are actually complexes of three loosely bound protein subunits:

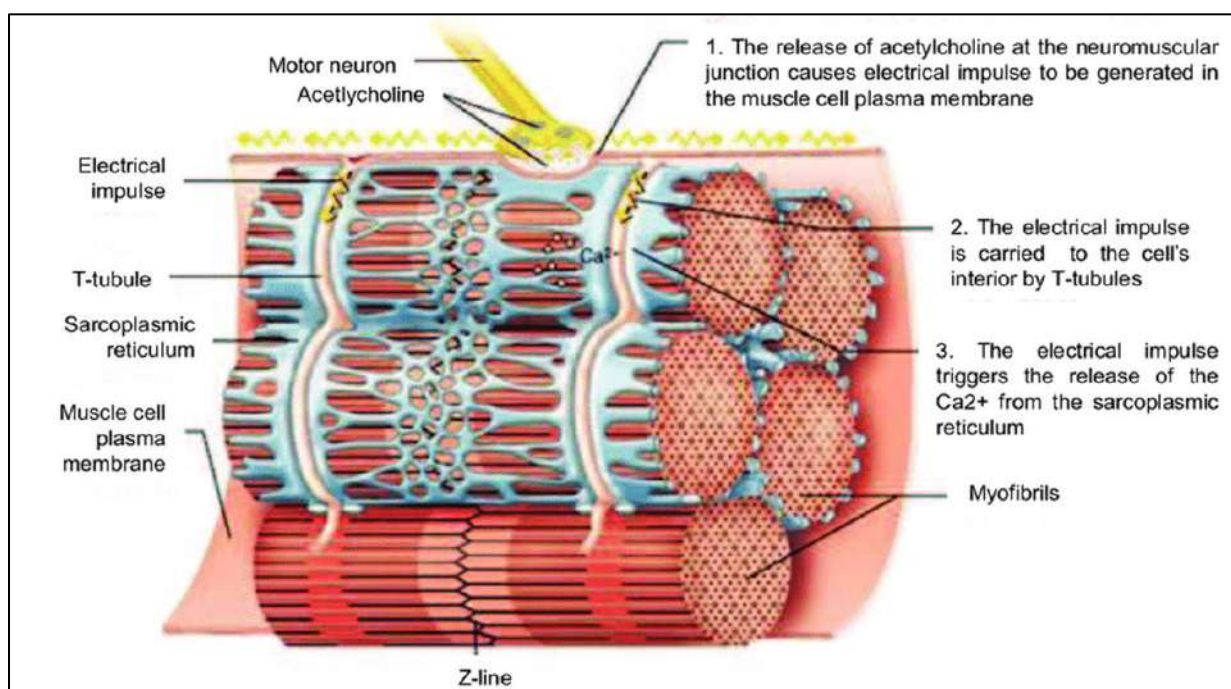
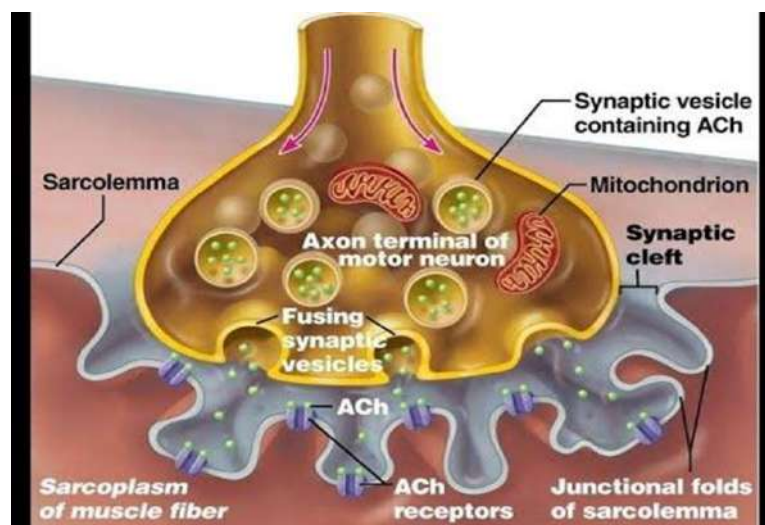
- **Troponin I:** has a strong affinity for actin
- **Troponin T:** has a strong affinity for tropomyosin
- **Troponin C:** has a strong affinity for calcium ions.

This complex (troponin I, T, C) is attach the tropomyosin to the actin. The strong affinity of the troponin for calcium ions initiate the contraction process.



Neuromuscular junction:

As the motor nerve reaches the muscle fiber, the axon terminal of motor neuron contains many small synaptic vesicles contain neurotransmitter acetylcholine which released in a synaptic cleft (a space between the axon terminal and the muscle fiber that transmits signals to muscle fiber). Ach binds with Ach receptors on the folds of the sarcolemma, then action potential occurs along the muscle fiber and the released Ca^{2+} ion from the endoplasmic reticulum will activate the muscle contraction.



What are the steps of muscle contraction? (The sliding filament theory)

➤ Inhibition of the actin filament by the troponin-tropomyosin complex.

In the relaxed muscle, the active sites on actin filament are inhibited or covered by the troponin tropomyosin complex, which represent the **relaxing proteins**, which inhibit interaction between actin and myosin.

➤ **Muscle activation:** Motor nerve stimulates the release of calcium ions into the muscle cell which activate muscle contraction.

➤ **Activation of the actin filament by calcium ions.**

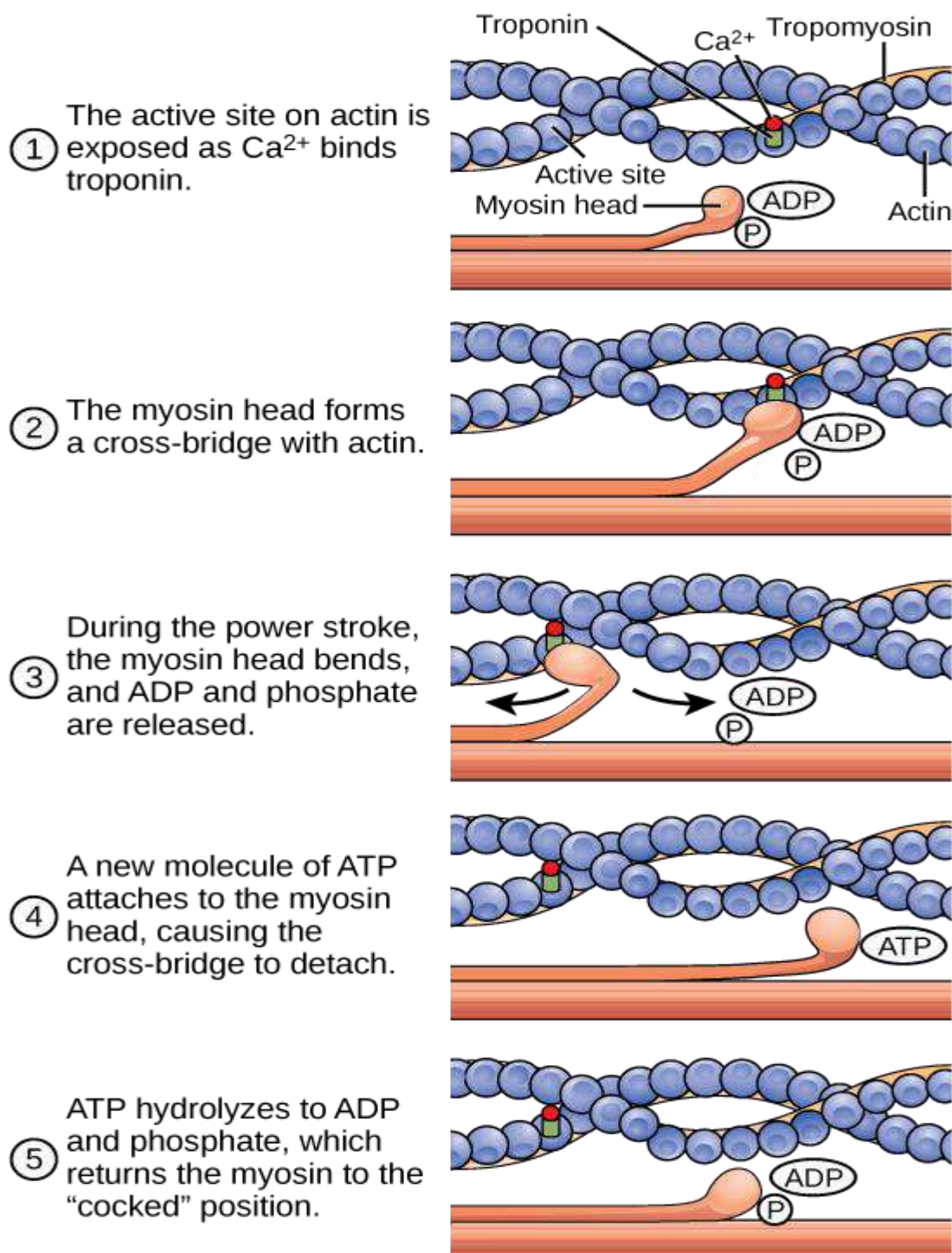
Calcium ions inhibit the effect of troponin-tropomyosin complex on actin filaments, by combining with troponin C, which leads to “uncovers” the active sites of the actin, thus allowing these active sites to attract the myosin cross-bridge heads and cause contraction to proceed.

➤ **Interaction of actin Filament and the myosin Cross-Bridges.**

As soon as the actin filament is activated by the calcium ions, the heads of the cross-bridges from the myosin filaments become attracted to the active sites of the actin filament, and this causes contraction to occur.

When a head of cross-bridges attaches to an active site of actin filaments, this attachment causes the head to tilt toward the arm and to drag the actin filament along with it. This tilt of the head is called the **power stroke**. After tilting, the head automatically breaks away from the active site. Next, the head returns to its extended direction. In this position, it combines with a new active site along the actin filament; the head then tilts again to cause a new power stroke, and the actin filament moves

another step. Thus, the heads of the cross-bridges bend back and forth and step by step walk along the actin filament, pulling the ends of two successive actin filaments toward the center of the myosin filaments.



Three sources of energy for muscle contraction:



- ✦ The first source of energy is **ATP**. The substance that carries a high-energy phosphate bond
- ✦ The second important source of energy is “**glycolysis**” of glycogen previously stored in the muscle cells.
- ✦ The third and final source of energy is **oxidative metabolism**, which means combining oxygen with the end products of glycolysis and with (carbohydrates, fats, and protein.) to liberate ATP.



Muscle Fatigue:

Fatigue results mainly from inability of the muscle fibers to generate force and performed the acquired activity, due to prolonged and strong contraction of muscles. It leads to damage in sarcolemma and sarcoplasmic reticulum, muscle exhaustion and pain.

Muscle fatigue may result from ATP depletion (depletion of muscle glycogen) or lack of acetylcholine in neuromuscular junction.





General Physiology

Lec.5

Nerve physiology



By- Ph.D Shaima'a A. Ahmed

Neuronal structure:

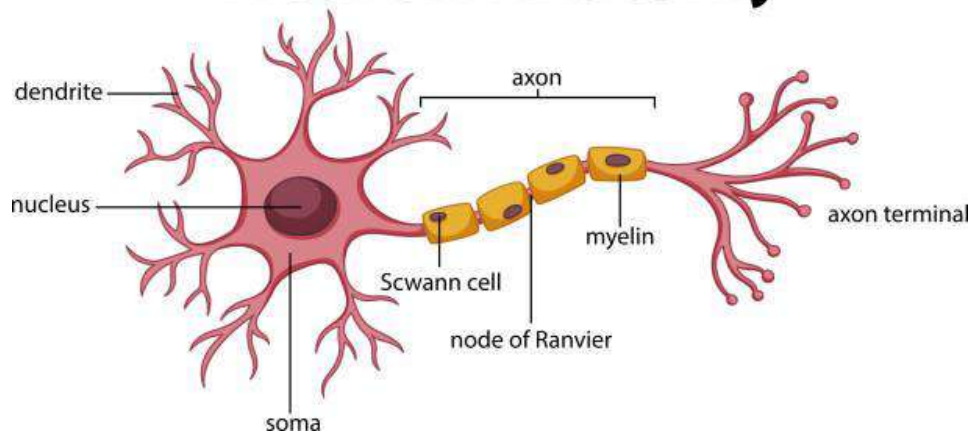
Neurons are the structural and functional units of the nervous system, that are able to respond to stimuli (such as touch, sound, light, and so on), conduct impulses, and communicate with each other (and with other types of cells like muscle cells) using electrochemical signals. The neurons in general are composed of 3 major parts:

- 1- **The soma (cell body of neuron):** is the location of the genetic material and synthetic activity of the neuron, contains specialized cytoplasm, single nucleus and other granules such as mitochondria and rough endoplasmic reticulum which is termed Nissl substance.
- 2- **Dendrites:** are shrub-like extensions from the cell body (soma) that receive incoming information from other neurons.
- 3- **Axon:** It starts from a thick area called the axon hillock which extends from the soma membrane to the periphery, conducts information collected by the dendrites and cell body away from the cell body by electrical signals called action potentials.

Axons can be very long and therefore facilitate transmission of information over large distances in the body. The axon is covered with a myelin sheath, a lipoprotein complex that insulates the axon and allows the electrical signal to travel much more quickly.

Myelin is interrupted by a small exposed area called “Node of Ranvier”
The combination of myelin and nodes allows action potentials to be conducted down the axon with much greater velocity than could be produced in an unmyelinated axon. Not all the nerve fibers are myelinated, some are not myelinated but surrounded by Schwann cells without the deposition of myelin.

Neuron Anatomy



Neuron function is to conduct an impulse (excitatory/ inhibitory).

Velocity of conduction: depends on:

- Myelination: the conduction velocity is much higher than that of unmyelinated nerve fibers
- Diameter: the conduction velocity increases with the diameter of nerve fiber.

The types of nerves in the body:

- **Autonomic nerves**, these nerves control the involuntary or partially voluntary activities of your body, including blood pressure, digestion, temperature regulation and urination

- **Motor nerves**, also known as efferent nerves, carry impulses away from the brain to muscles and gland
- **Sensory nerves**, also known as afferent nerves, carry impulses from sensory receptors towards the brain. for example from the mechanoreceptors in skin.

Types of neural communication:

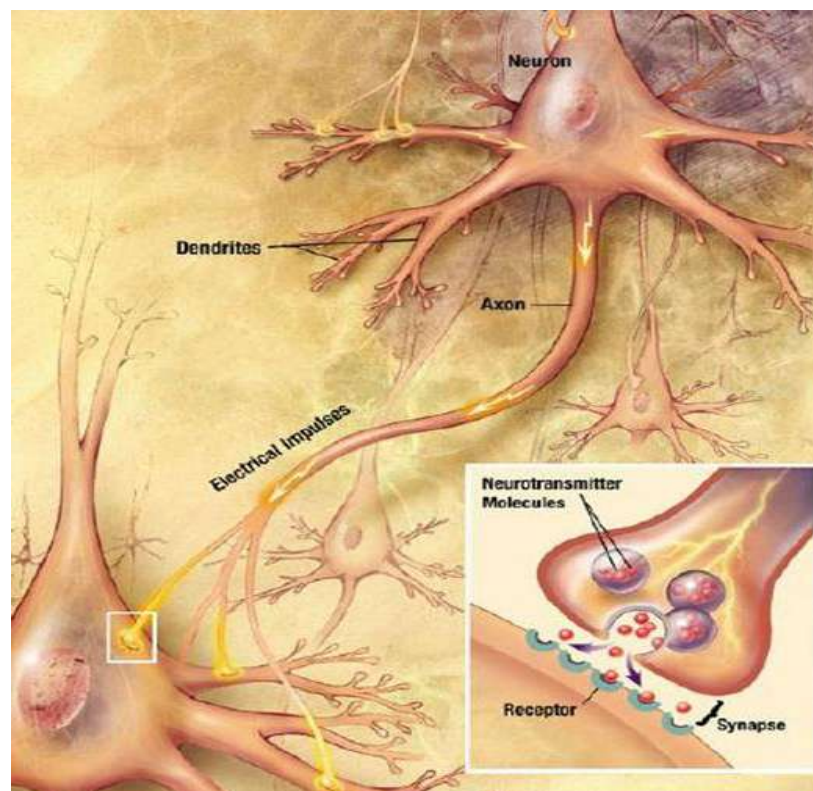
The neurons communicate with each other by two types of communication; both types are physiochemical disturbances due to change in conduction across the cell membrane:

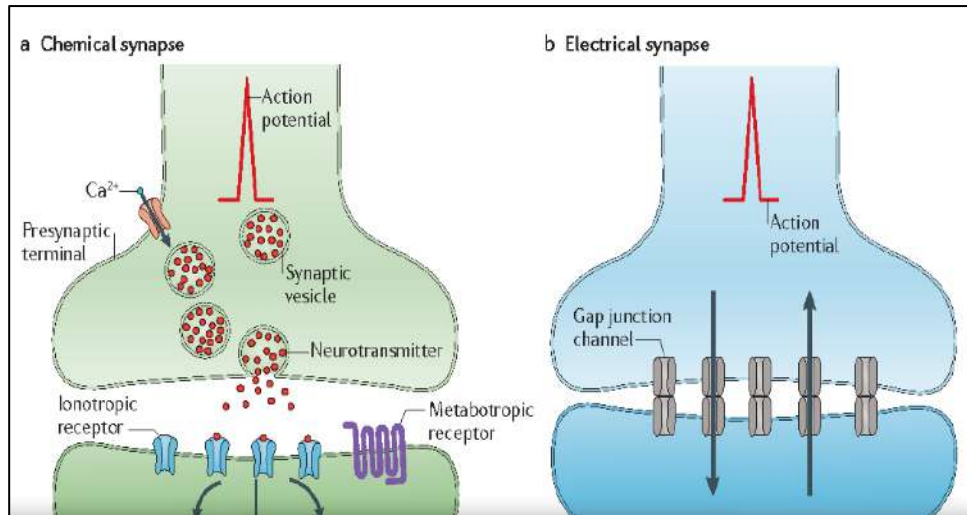
- The action potential (**nerve impulse**): is a propagated disturbance of electrical charge that travels along the membrane of a neuron to send information for long distances without any loss of energy. It occurs because of a difference in electrical charge (ions) across the plasma membrane of a neuron by chemical signals from a nearby cell
- The electronic potential (**synaptic transmission**): The place where an axon terminal meets another cell is called a **synapse**. It is a local non-propagated potential used for communication between neurons which are very close to each other where the transmission of a nerve impulse occurs. The cell that sends the nerve impulse is called the presynaptic cell, and the cell that receives the nerve impulse is called the postsynaptic cell.

Synaptic transmission:

Neurons communicate at synapses. Two types of synapses have been identified: electrical and chemical.

- Electrical synapses are the passage of ions from cell to another by a gap-junction, this type of synapses uncommon in the adult mammalian nervous system. They are more common in the embryonic nervous system, and they also important for cell-to-cell communication in smooth and cardiac muscle
- Chemical synapses: The structure of a chemical synapse includes the terminal portion of the axon, called the axon terminal, a synaptic cleft, and a group of receptors in the postsynaptic cell membrane .The axon terminal contains vesicles filled with chemical neurotransmitter molecules. These molecules are released into the synaptic cleft when an action potential enters the axon terminal.





Types of neurotransmitters:

Neurotransmitters are chemical messengers that are used by the nervous system to transmit messages between neurons, or from neurons to muscles, by converting the electrical signals from axons to chemical ones causing specific response.

The below table demonstrate some types of neurotransmitters and their functions:

Neurotransmitters	Function
1. acetylcholine	Plays a major role in controlling movement, cognitions and autonomic control.
2. glutamate	General excitation and transmission of sensory information into the CNS
3. serotonin	Involved in functions such as sleep, memory, mood and others. It is also produced in the gastrointestinal tract in response to food.
4. Glycine and GABA (gama-aminobutyric acid)	General inhibitory neurotransmitters.

Reflex action:

A reflex, is an involuntary and rapid movement in response to a stimulus, the pathway traveled by the nerve impulses during a reflex is called 'reflex arc' which involved two types: **autonomic** reflex arc (inner organs) and **somatic** reflex arc (muscles)

Example of reflex is **the pupillary light reflex**. If a light is flashed near one eye, the pupils of both eyes contract. Light is the stimulus; impulses reach the brain via the optic nerve; and the response is conveyed to the pupillary musculature by autonomic nerves that supply the eye

GOOD LUCK

Ph.D Shaima'a A. Aljubouri



General Physiology

Lec.6



Neuron action potential

By- Dr. Shaimaa A. Ahmed

Introduction:

Electrical potentials exist across the membranes of virtually all cells of the body. Some cells, such as nerve and muscle cells, generate rapidly changing electrochemical impulses at their membranes, and these impulses are used to transmit signals along the nerve or muscle membranes.

Neural action potential:

It is a rapid change in the membrane potential (ions) that spread along the nerve fiber membrane. Each action potential begins with a sudden change from the normal resting negative membrane potential (typically -70 mV) to a positive potential (typically about $+30$ mV) and ends with rapid change back to the negative potential in a very short period of time (just a few milliseconds). Action potential occurs due to the role of three factors:

- ◆ Diffusion of potassium ions. (passive)
- ◆ Diffusion of sodium ions. (passive)
- ◆ The contribution of Na-K ion pump. (active)



Sodium channel



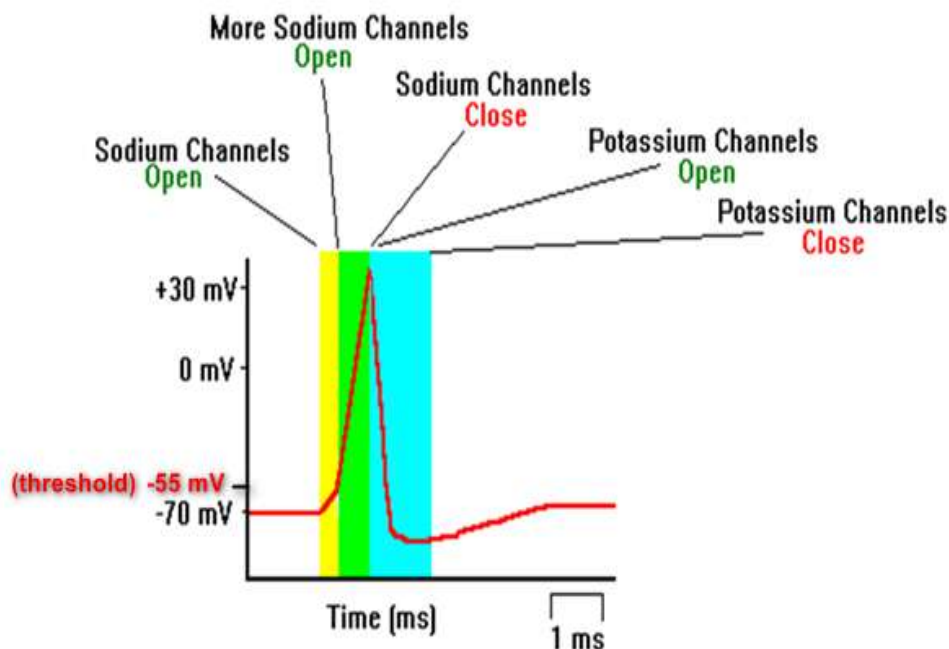
Potassium channel



Na⁺ - K⁺ pump

How does the neural action potential occurs?

The stimulus causes the sodium gates to open and because there's more sodium on the outside than the inside of the membrane (voltage value **-70mV** which represent the resting state) sodium then diffuses ,here the potential become less negative value **-55mV**, which called **threshold potential** or **firing level** (The minimum stimulus needed to achieve an action potential). After that sodium more and rapidly enter the nerve cell, causes the membrane potential to become positive (**+30 mV**) called **Action potential**. The sodium channels then close again. The potassium channels open and positively-charged potassium ions diffuse out. As these positive ions go out, the inside of the membrane once again becomes negative with respect to the outside.



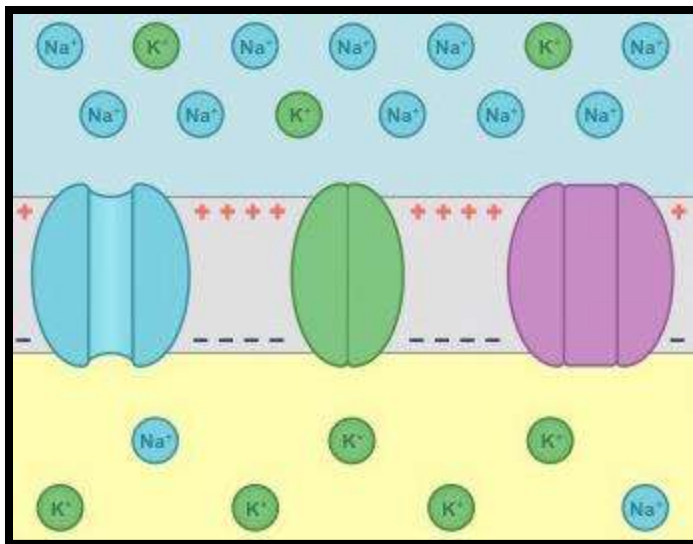
The main stages in action potential are:

➤ Resting stage of neurons:

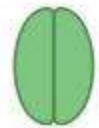
The resting stage is the resting membrane potential before the action potential begins. The membrane is said to be “polarized” during this stage. It varies depending on the cell type, between -70 mV (in small nerve fibers) and -90 mV (in large nerve fibers). During resting potential there are:

- ◆ More sodium ions outside than inside the neuron
- ◆ More potassium ions inside than outside the neuron
- ◆ Proteins with negative charges inside the cell.

The net result polarization is negative inside and positive outside the cell.



Sodium channel



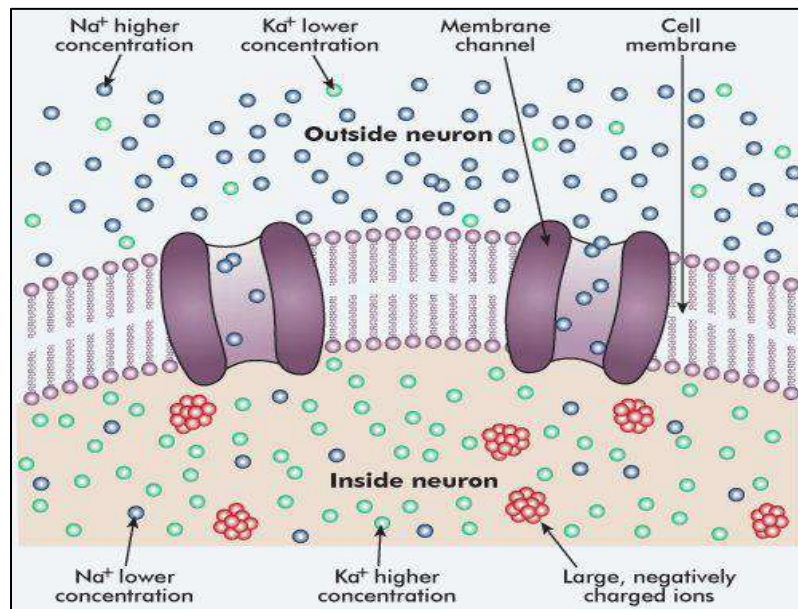
Potassium channel



Na⁺ - K⁺ pump

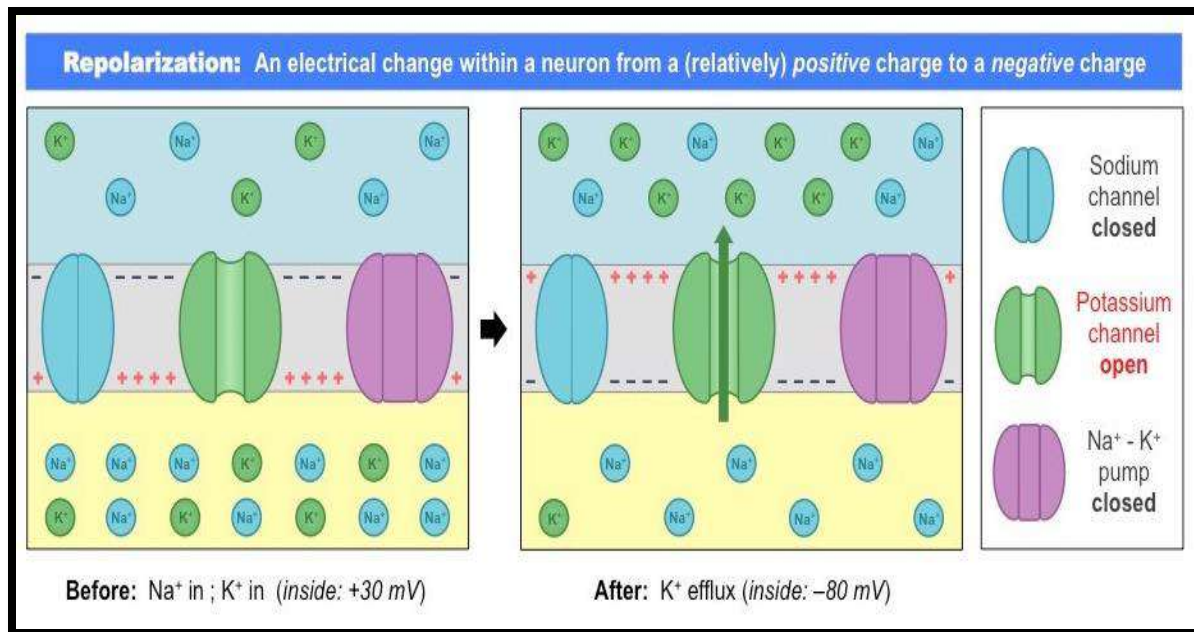
➤ Depolarization Stage:

At this stage, the membrane suddenly becomes permeable to sodium ions because of the activation of **all** Na^+ channels, allowing tremendous numbers of sodium ions to diffuse to the interior of the axon. The normal “polarized” state of -70 millivolts is rising rapidly in the positive direction until it reaches the $(+35\text{mV})$.



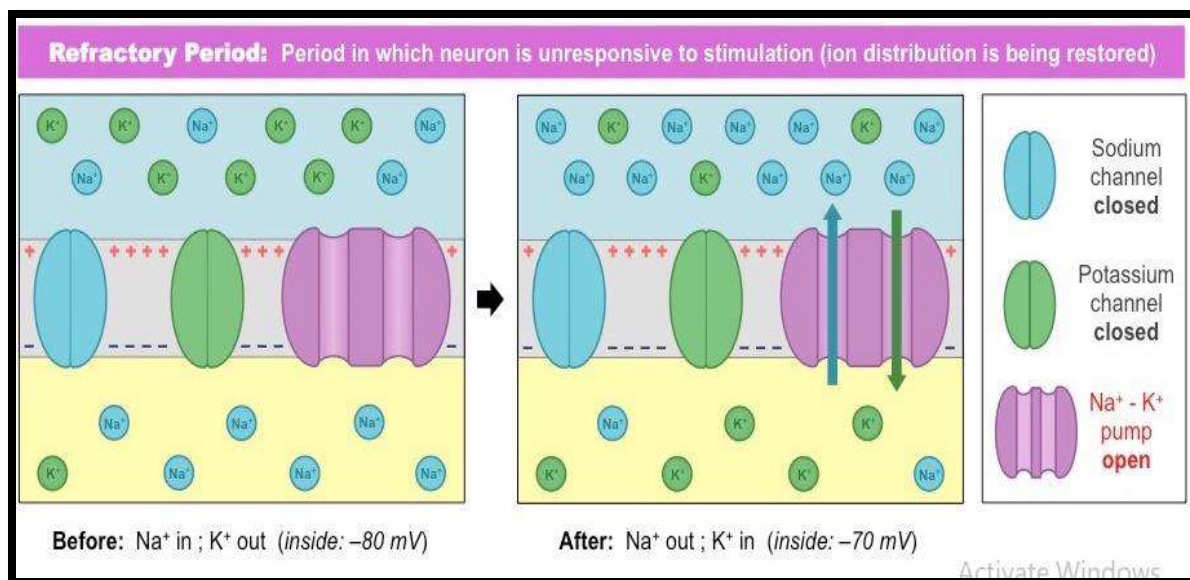
➤ Repolarization Stage:

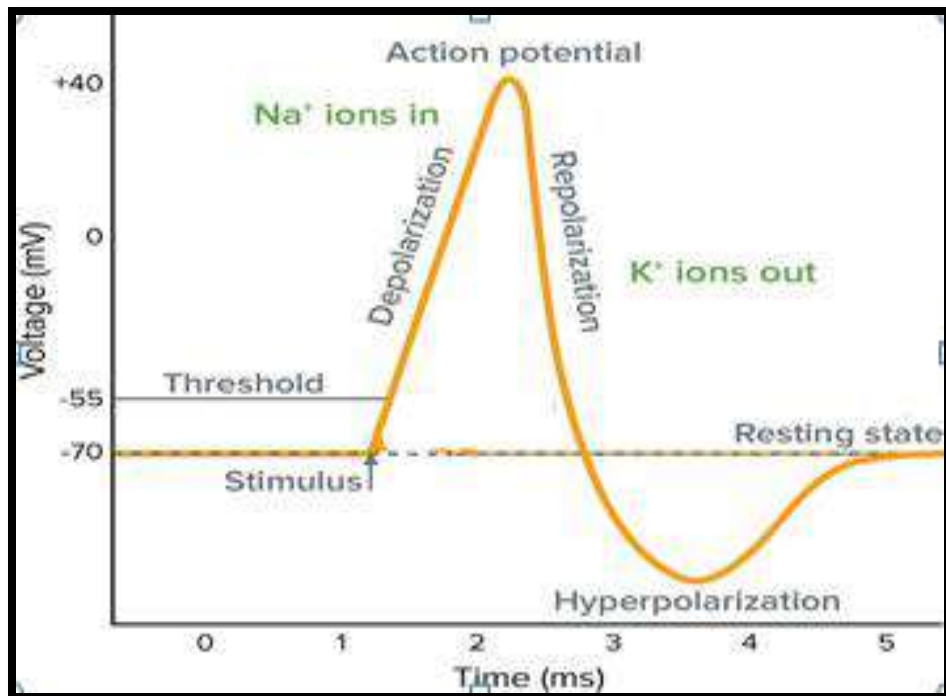
After the membrane becomes highly permeable to sodium ions, the sodium channels begin to close and the potassium channels open. Then, rapid diffusion of potassium ions to the exterior re-establishes the normal negative resting membrane potential.



➤ Hyperpolarization stage:

At this stage the cell being more negative than its typical resting membrane potential. As the potassium channels stay open and continue to exit ions out of the neuron. When potassium channels close, the sodium-potassium pump works to reestablish the resting state by actively transporting the K^+ ions inside the cell and Na^+ ions outside the cell to reach the resting membrane potential (polarized state -70mV).





Good Luck

Dr. Shaimaa



General physiology

Lec. 7

Blood Physiology

قسم تقنيات التخدير

المرحلة الأولى

م.د. شيماء عبدالله احمد



Blood:



Blood is a dynamic and complex living tissue, which circulate in a closed system of blood vessels, carrying oxygen and nutrients to every part of the body and transporting carbon dioxide and other waste products to the lungs, kidneys, and liver for disposal. Whole blood is bright red in color, which derives from the oxygenated iron in hemoglobin and it can be seen in arteries blood samples. Deoxygenated blood is darker red, which can be seen in venous blood samples.

Facts about the blood:



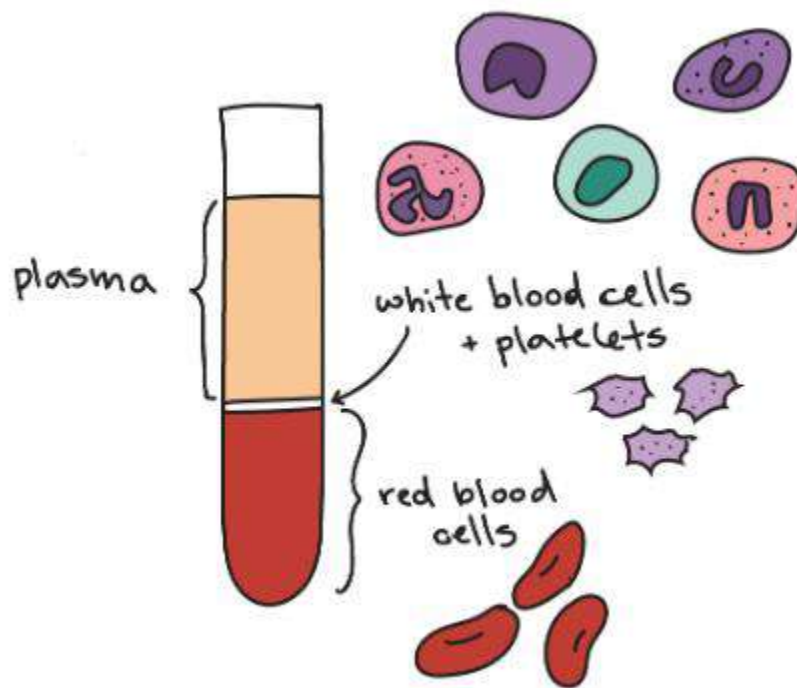
- ✗ Typical volume in adult male 5-6 liters and in adult female 4-5 liters.
- ✗ PH range from 7.35 to 7.45 (slightly alkaline).
- ✗ Blood accounts for 6% to 8% of the body weight of a healthy adult.
- ✗ Slightly warmer than body temperature 38 °.
- ✗ Blood is a connective tissue.
- ✗ Much more dense than pure water.



Major components of the whole blood:

Whole blood like other connective tissue composed of formed elements and intercellular matter. The formed elements are **red** and **white** blood cells and cell fragments called **platelets**. The intercellular matter is the liquid component of blood (**plasma**).

As different blood components have different relative density, sediment rate and size they can be separated when centrifugal force is applied. In increasing order, the specific gravity of blood components is plasma, platelets, leucocytes (Buffy Coat) and packed red blood cells.



Blood functions:

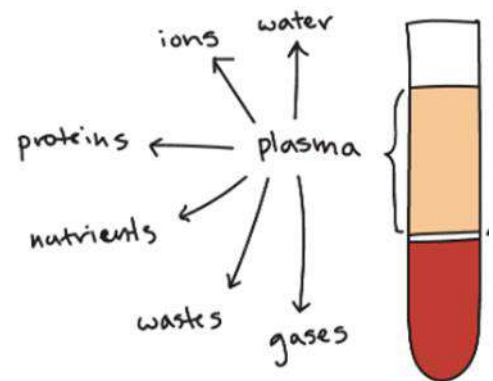


Blood is carrying out four major roles:

- **Transport:** Blood carries important substances from one area of the body to another, including antibodies, ions, vitamins, hormones, lipids and minerals. For example O₂ and CO₂ bound to hemoglobin within red blood cells. In addition to transporting materials, blood also transports heat by distributing heat throughout the body.
- **Hemostasis:** (the arrest of bleeding). The failure to stop bleeding after injury is called hemorrhage and can quickly lead to bleeding to death.
- **Homeostasis:** It is a steady state of cell function, the blood system plays a crucial role in preserving homeostasis by maintaining pH, ion concentrations, osmolality, temperature and nutrient supply.
- **Immunity:** Blood leukocytes are involved in the body's battle against infection by microorganisms, the blood's defense system is efficient enough to eliminate the pathogens or to prevent their spreading before they can cause substantial bodily harm.

Plasma:

About 55% of whole blood is made up of plasma which is the liquid portion of the blood. It has a cloudy, pale yellow color and comprises mostly water (93%), with the remaining (7%) being various dissolved solutes that would be organic



substances (proteins, fatty acids, glucose, metabolic waste, gases ect.) and inorganic substances (electrolytes as Na^+ , Ca^{++} , Mg^{++} , Cl^- , HCO_3 and others).

Plasma proteins:

Proteins are one of the main dissolved organic solutes of plasma; there are three types of plasma proteins:

Albumins: a small protein produced by the liver, it account for about 60% of total plasma proteins concentration. Albumin plays a role in osmotic pressure and in transport fatty acids, hormones, drugs, and other substances through blood

Globulins: constitute about 36% of the total plasma proteins. The globulins type (α and β) are produced in the liver and perform functions as a transporters (thyroxin) or as a substrates (plasminogen). γ - Globulins, produced by lymphoid tissue, are antibodies necessary for immune defense.

Fibrinogens: are large molecules synthesized in the liver, which account for about 4% of the total plasma protein concentration. Fibrinogen contributes to the clotting of blood when it converted to fibrin.

Other dissolved solutes in the plasma are lipids (cholesterol, phospholipids, and triglycerides), glucose, gases (O_2 , CO_2) and metabolic waste (as urea, bilirubin, creatinine...)

Serum:

When blood is allowed to clot or coagulate, usually at room temperature for 15 to 30 minutes, the remaining fluid after centrifugation is referred to as serum, that is a clear, watery fluid which has the same composition as plasma except the clotting factors (fibrinogen) and blood cells.

The key differences between plasma and serum are:

- Plasma contains fibrinogen which is absent in serum.
- Serum is obtained after the clotting of blood, while plasma can be obtained before the coagulation of the blood.
- Serum is mostly used for blood typing but is also used for diagnostic testing. Plasma, on the other hand, is mostly used for blood-clotting related problems.

تمنياتي لكم بالتوفيق

د.شيماء عبدالله احمد





General Physiology

Lec.8

Red blood cells

قسم تقنيات التخدير

المرحلة الاولى

2020-2012

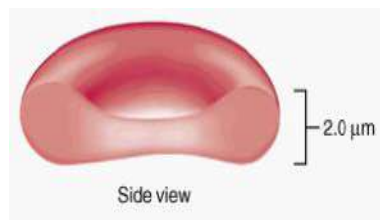
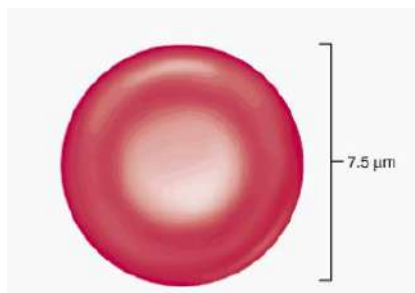


BY- Dr.Shaymaa A. Ahmed

Red blood cells (also referred to as *erythrocytes*) are the most common cell type in the blood and are the principle means of delivering oxygen to cells via the circulatory system.

Shape and size:

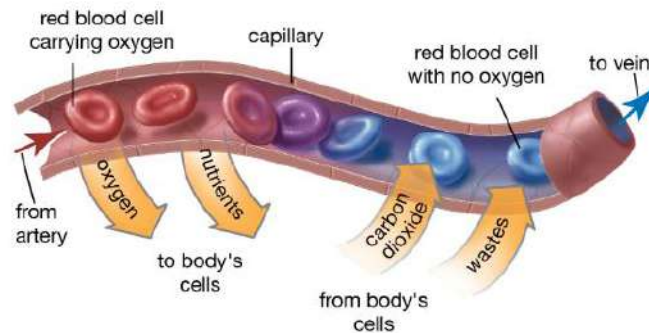
In humans, mature red blood cells are flexible biconcave disks with a diameter of about 7 μm and a thickness of 2.5 μm . red blood cell count as cells per cubic millimeter: (5-5.8 million) in adult male and (4-5.2 million) in adult female. Mature RBCs lack a nucleus and most organelles.



Functions:

- ✚ A major function of RBCs is to transport hemoglobin, which, in turn, carries oxygen from the lungs to the tissues.
- ✚ Erythrocytes also play a major role in the homeostasis of blood pH. The RBCs contain a large quantity of carbonic anhydrase, an enzyme that have a major role in transporting CO₂ in the form of bicarbonate

ion (HCO_3^-) from the tissues to the lungs where it is reconverted to CO_2 and expelled into the atmosphere as a body waste product.

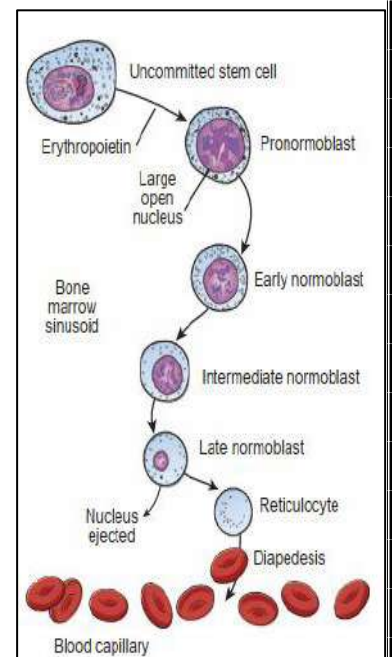


Production sites:

In the early weeks of embryonic life, primitive, nucleated RBCs are produced in the yolk sac. During the middle of gestation, the liver, spleen and lymph nodes are the sites of RBC production. After birth, RBCs are produced exclusively in the bone marrow.

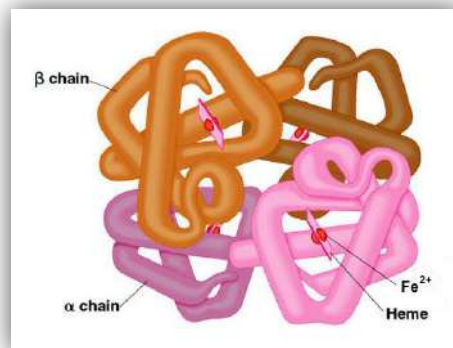
Origin and formation:

Erythropoiesis is the process by which red blood cells are produced. It is stimulated by decreased O_2 delivery to the kidneys which then secrete the hormone *erythropoietin* that regulates the differentiation of the uncommitted stem cell forming erythroblasts or called normoblasts (early, intermediate and late normoblasts), reticulocytes, and finally, mature erythrocytes, which enter the bloodstream. A common symptom of patients with chronic kidney disease is anemia from the lack of erythropoietin. Erythrocytes circulate for about 120 days before they are destroyed by macrophages.



Hemoglobin:

Each red blood cell contains several hundred of Hemoglobin molecules which is an oxygen-carrying protein pigment in the red blood cells of vertebrates. It is a globular protein with a molecular weight of 64,500 daltons, consists of a globin portion and four heme groups. This complex protein possesses four polypeptide chains: two α -globin molecules and two molecules of another type of globin chain (β , γ , δ).



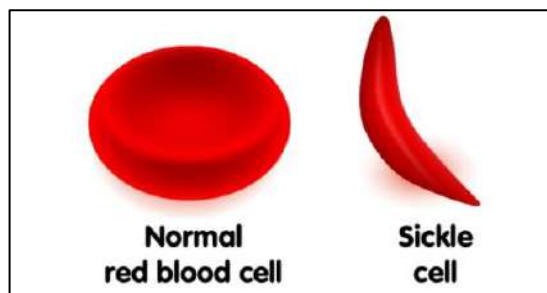
Hemoglobin types:

There are many types of hemoglobin (normal and abnormal types). The normal types of hemoglobin include:

- ✿ **Hemoglobin A:** the most common type of hemoglobin in healthy adults. consists of two α polypeptide chains and two β polypeptide chains ($\alpha_2\beta_2$).
- ✿ **Hemoglobin F:** fetal hemoglobin. The major type in infants and newborns, consist of ($2\alpha\ 2\gamma$) polypeptide chains and it is replaced by HgA shortly after birth. Its levels in circulating blood cells decrease rapidly during infancy and reach a concentration of 0.5% in adults.

The abnormal types of hemoglobin are caused by mutations occur in genes that controlled the globin chains, this has resulted in the production of many types of abnormal Hb molecules include:

✿ **Hemoglobin S:** or called sickle cell hemoglobin (HbS) which found in sickle cell disease, an inherited disorder that causes the body to make sickle-shaped red blood cells which can get stuck in the blood vessels, causing severe and chronic pain, infections, and other complications. HbS differs from normal adult HbA because of the substitution of a single amino acid in each of the two β chains.



Anemias:

Anemia means deficiency of hemoglobin in the blood, which can be caused by either too few RBCs or too little hemoglobin in the cells. Some types of anemia and their physiological causes are described in the following sections.

✿ **Blood loss anemia:** after rapid and chronic hemorrhage.

✿ **Aplastic Anemia:** Due to bone marrow dysfunction especially after exposure to high dose radiation or chemotherapy for cancer treatment or high doses of certain toxic chemicals can damage stem cells of the bone marrow followed by anemia

- ✿ **Megaloblastic anemia:** due to loss of vitamin B12, folic acid, and intrinsic factor from the stomach mucosa (it has an important role in absorption of vitamin B12 in the intestine).
- ✿ **Hemolytic anemia:** due to different abnormalities of the RBCs, many of which are hereditarily acquired, make the cells fragile, so they rupture easily as they go through the capillaries. The life span of the fragile RBC is so short that the cells are destroyed faster than they can be formed and serious anemia results.

Polycythemia:

Means the production of large quantities of RBCs by the blood forming organs and the RBC count rises to 6-8 million/mm³, and that occurs as the oxygen concentration in the tissues becomes too low (hypoxic) such as high altitude and cardiac failure (**physiological polycythemia**). Pathological conditions of polycythemia known as **polycythemia Vera** is caused by a genetic defect in the blast cells that produce the blood cells. The blast cells no longer stop producing RBCs when too many cells are already present.

Good Luck





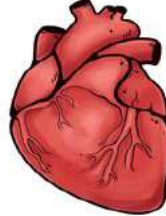
General Physiology Lec. 11

قسم تقنيات التخدير
المرحلة الاولى
2020-2021



Heart Physiology

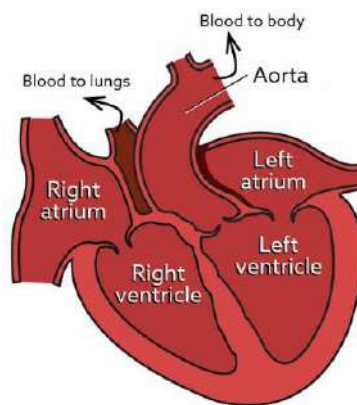
By-Dr.Shaymaa A. Ahmed



Introduction:

Heart is a hollow muscular organ that pumps the blood throughout the body. It contracts and relaxes in a regular repeating cycle to pump blood. The period of time the heart spends in contraction is called **systole**, and the time it spends in relaxation is called **diastole**.

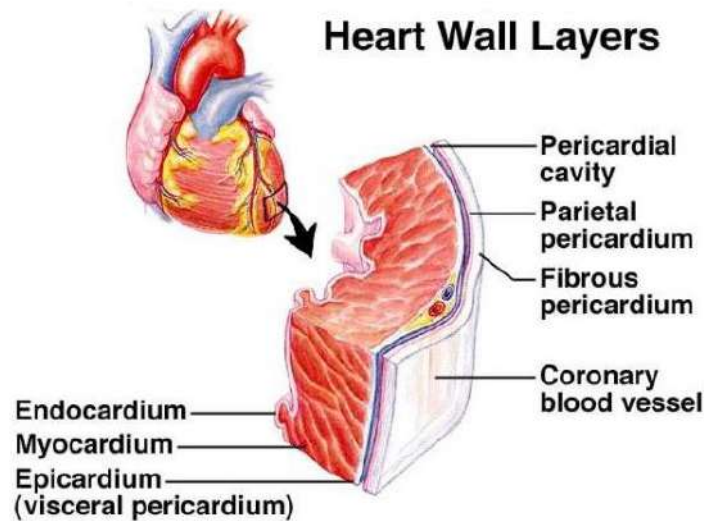
The heart is actually two separate pumps: a *right heart* (composed of *atrium* and a *ventricle*) that pumps blood through the lungs, and a *left heart* (composed of *atrium* and a *ventricle*) that pumps blood through the systemic circulation that provides blood flow to the other organs and tissues of the body. Each atrium pumps the blood, helping to move it into the ventricle. The ventricles then pump the blood either (1) through the pulmonary circulation by the right ventricle or (2) through the systemic circulation by the left ventricle.



Heart wall:

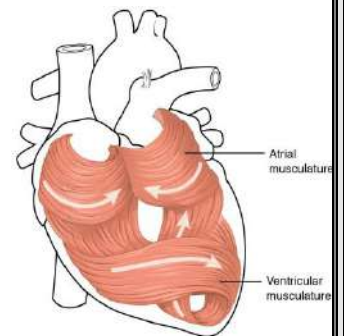
Composed of 4 layers, from inside it is:-

- ✿ Endocardium- consists of the endothelial cells. Forming a smooth membrane.
- ✿ Myocardium - is the muscular tissue of the heart.
- ✿ Epicardium – it consists of a thin layer of connective tissue and fat.
- ✿ Pericardium - is the thick, membranous sac that surrounds and protect the heart.it consist of two layers: fibrous pericardium and parietal pericardium.



Heart muscles:

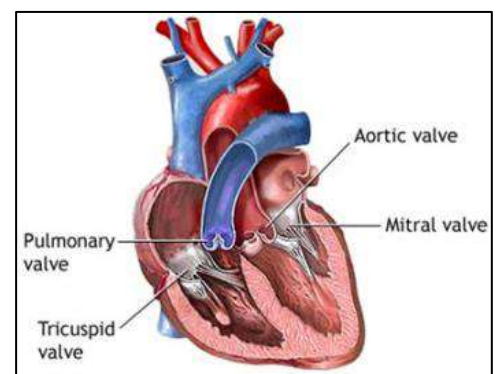
The heart is composed of three major types of cardiac muscle: *atrial muscle*, *ventricular muscle*, and specialized excitatory and conductive muscle fibers that help spread of the impulse (action potential) rapidly throughout the heart causing a contraction.



Heart valves:

Valves are thin membranous cusps that allow blood to flow in only one direction through the heart. Heart valves separate the atria from the ventricles, or the ventricles from a blood vessel. The main types of valves found in mammals are:

- ✿ The atrioventricular (AV) valves, that include:
 1. Mitral valve (bicuspid valve) located between the left atrium and the left ventricle
 2. Tricuspid valve, located between the right atrium and the right ventricle.



The (AV) valves allow blood to flow from the atrium to the ventricle but prevent backward flow from the ventricle to the atria.

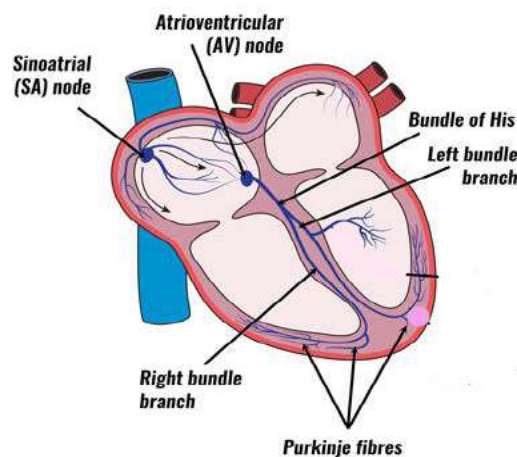
- ✿ The semilunar (SL) valves, that include:
 3. Aortic valve, located between the left ventricle and the aorta.
 4. Pulmonary valve, located between the right ventricle and the pulmonary artery.

The pulmonary and aortic valves allow blood to flow into the arteries during ventricular contraction (systole) but prevent blood from moving in the opposite direction during ventricular relaxation (diastole).

Heart conductive system:

The main parts of the system are:

- ✿ The sinus node or **SA node**: also called sinoatrial node, located in the superior lateral wall of the right atrium. The excitation signal (impulse) is created by this node and spreads across the through myocardium of right and left **atria**, causing them to contract, so it is called the pacemaker of the heart. It's also quickly transmitted to the atrioventricular node.
- ✿ The **atrioventricular node** – After the electrical impulses spread across the atria, they converge at this node which located within the atrioventricular septum, near the opening of the coronary sinus, at which impulse from the atria is delayed before passing into the ventricles, and then passes from the atrioventricular node into the atrioventricular bundle.
- ✿ Atrioventricular Bundle (**bundle of HIS**): specialized tissue of the **AV node**, and serves to transmit the electrical impulse from the AV node to the Purkinje fibers of the ventricles. dividing into two main bundles:
 - Right bundle branch – conducts the impulse to the right ventricle
 - Left bundle branch – conducts the impulse to the left ventricle.
- ✿ **Purkinje Fibers**: specialized cells that are able to rapidly transmit cardiac action potentials from the atrioventricular bundle to the myocardium of the ventricles.

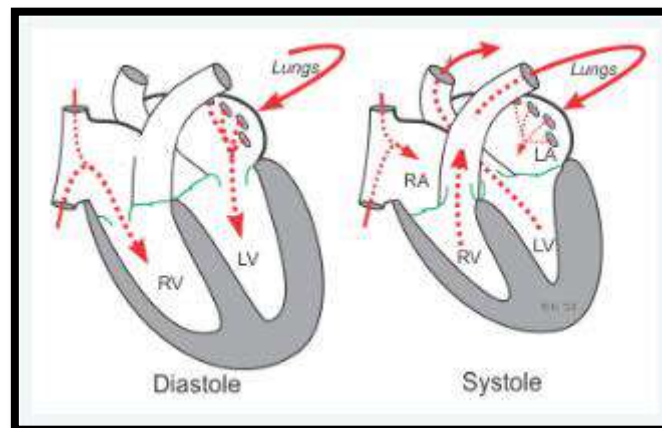


Cardiac cycle:

The events that occur from the beginning of one heartbeat to the beginning of the next are called the *cardiac cycle*. A single cycle of cardiac activity can be divided into two basic phases - a period of relaxation (not contracting) called **diastole**, during which the heart fills with blood, followed by a period of contraction called **systole**.

During diastole the ventricles are relaxed (not contracting), blood is flow passively from the atria into the ventricles through atrioventricular valves (mitral and tricuspid). At the end of diastole, both atria contract which propels an additional amount of blood into the ventricles.

During systole the ventricles contract and eject blood into the aorta and pulmonary artery. At this phase, the atrioventricular valves are closed; therefore no blood is entering the ventricles.



**trust yourself
and
move forward**



Dr.Shaymaa