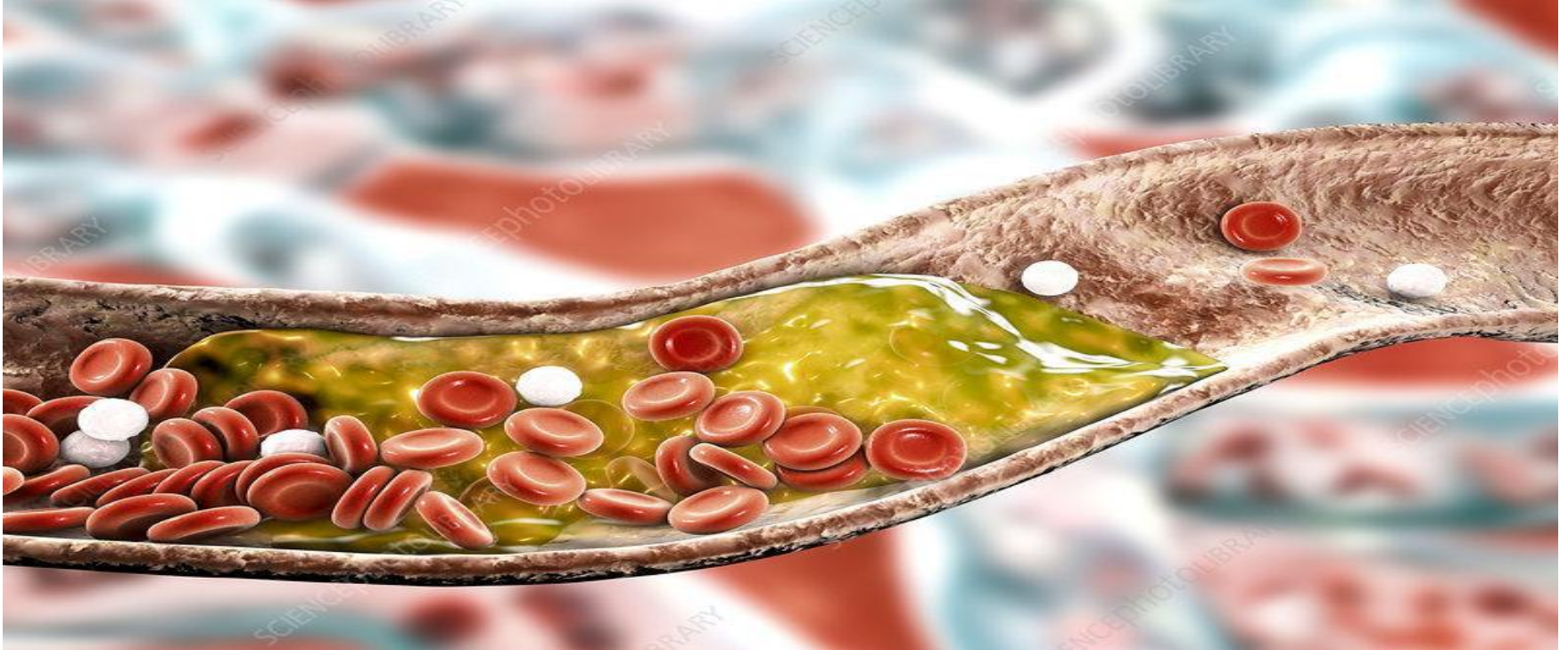


BLOOD



By

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LEARNING OUTCOMES

- ✓ Recognize the components of blood.
- ✓ Explain blood film preparation to study different blood elements.
- ✓ Morphology identification of red blood cells ,platelets, and different types of white blood cells by LM &EM.
- ✓ Identify the function of different blood elements.

➤ **Blood** is a special type of connective tissue.

➤ Mesenchymal in origin

➤ Blood composed of **formed blood elements** (represent the cells) suspended in fluid extracellular material called **plasma** (represent the matrix).

➤ **Blood cells** derived from stem cells in bone marrow.

➤ **Plasma protein** formed in liver.

➤ It is about 5 L of blood in an average adult.

Components of Blood

Blood cells
45%

- 1- Red blood cells (corpuscles) or erythrocytes (about 44%).**
- 2- White blood cells (1%) or leukocytes.**
- 3- Platelets (less than 1%).**

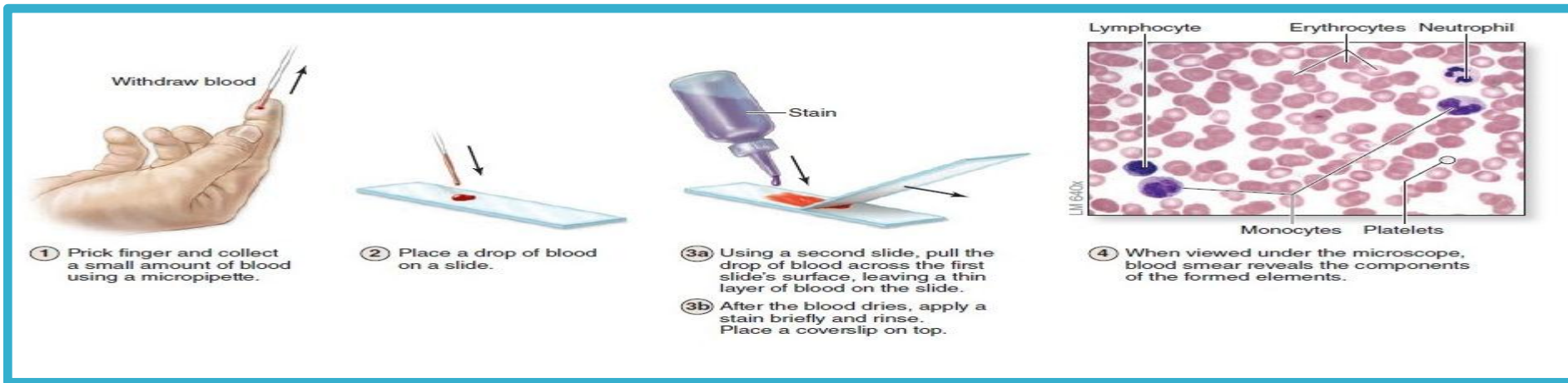
Blood plasma
55%

It is a *yellow* fluid in which the blood cells are suspended. It composed of:

- 1- Water 90 %.**
- 2- Plasma proteins 7% (albumin, globulin, fibrinogen and prothrombin) .**
- 3- inorganic salts and organic substances (amino acids, glucose, hormones, and enzymes**

How we can study blood elements

By preparing Blood film or peripheral blood smear to demonstrate the blood cells. Blood film It is a thin layer of blood on a glass microscopic slide to be stained to allow the various blood cells to be examined microscopically.



Blood Stain: Giemsa stain or Leishman stain used to differentiate blood cells by properties of their **nuclei and cytoplasmic granules**. It composed of mixture of:

- ✓ Eosin (acidic stain) → stain basic cellular components (**pink**).
- ✓ Methylene blue (basic stain) → stain acidic cellular components (**violet**).

When methylene blue is oxidized, it is converted to azures.

Cell components as lysosomes are stain **reddish blue** by binding to azures.

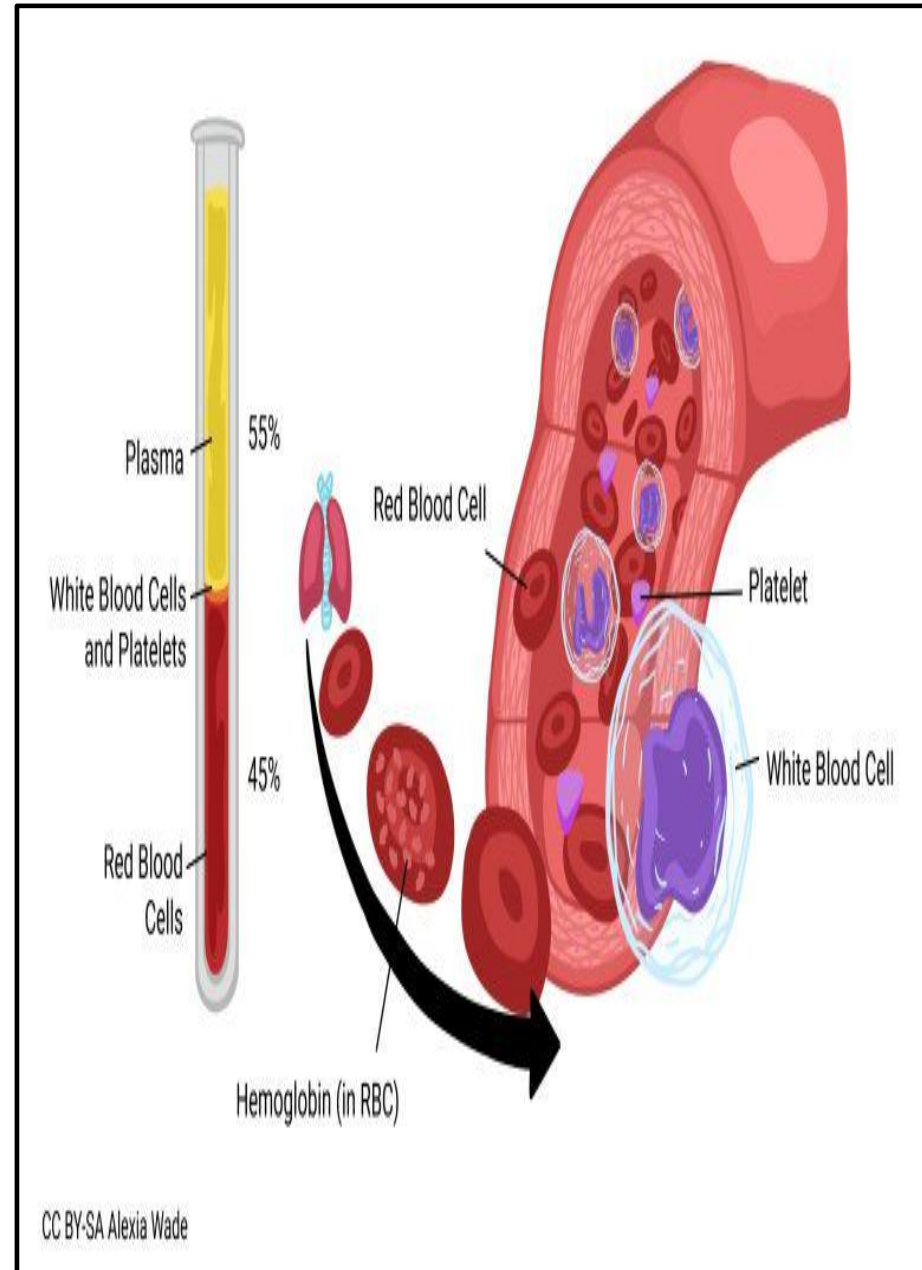
Blood cells

1-True: have nucleus.

- **White blood cells** or leukocytes (1%).

2-Not True:

- Red blood corpuscles or erythrocytes (44%).
- Platelets or thrombocytes (less than 1%).



Erythrocytes

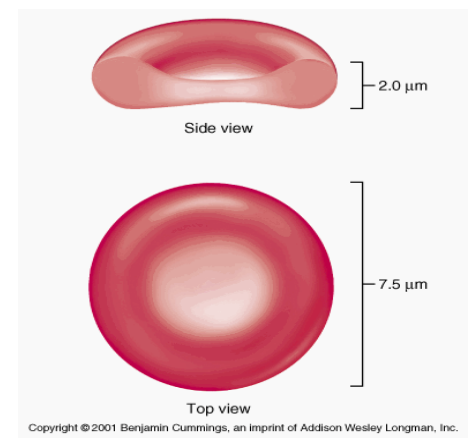
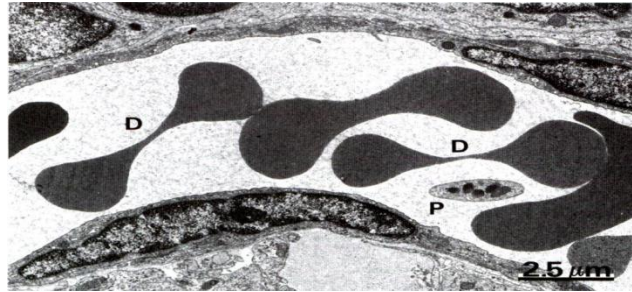
Red blood cells (RBCs)

- **Erythrocytes** (red blood cells or RBCs) are lacking nuclei and completely filled with the O₂ carrying protein **hemoglobin**.
- RBCs are the only blood cells whose function does not require them to leave the vasculature.

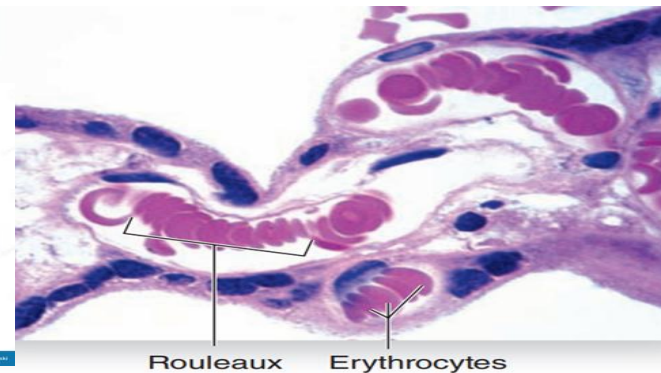
□ Number of RBCs

- Males: 5-5.5 million / cubic millimeter of blood
- Females: 4.5-5 millions / cubic millimeter of blood

□ Shape : Rounded biconcave discs



- They are soft and quite flexible and can change their shape *to bend* and adapt to the irregular turns and pass through small capillaries.
- They adhere together in stagnant blood due to their surface tension forming an arrangement resembling **stacked coins** called **rouleaux appearance**



Size of RBCs:

The normal diameter of RBCs varies between 6 - 9 microns (7.5 μm average).

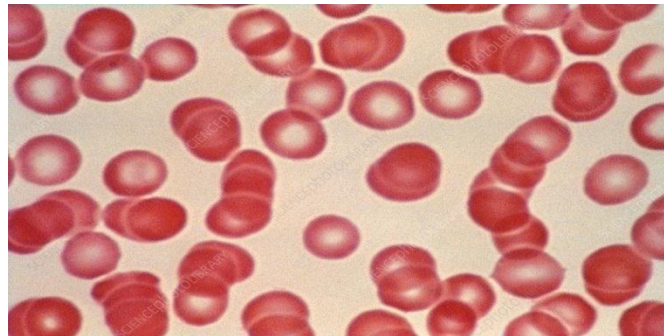
Life Span:

- RBCs survive in peripheral blood for about **120 days**.
- Old and deformed RCBs are removed by **macrophages** in the spleen and bone marrow.

Colour: depends on their *hemoglobin (Hb) content*.

Unstained erythrocytes have greenish yellow colour

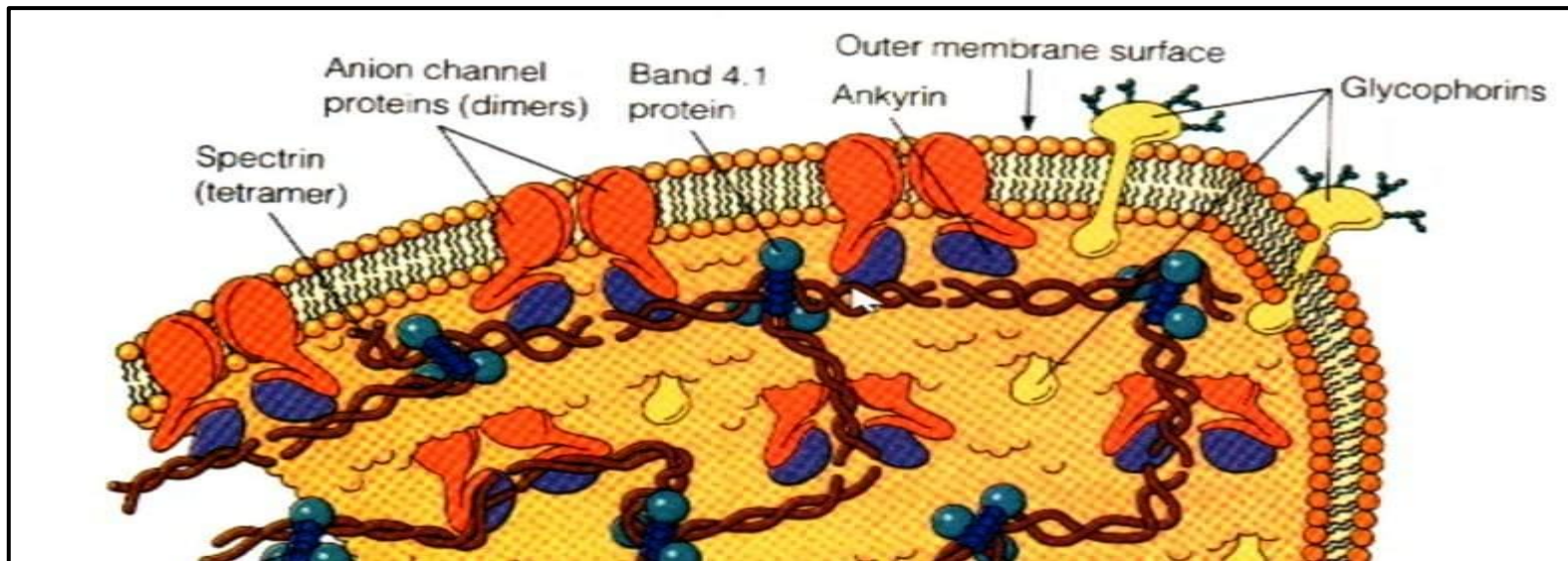
in stained blood films: deep pink in colour (**eosinophilic**).



□ Structure:

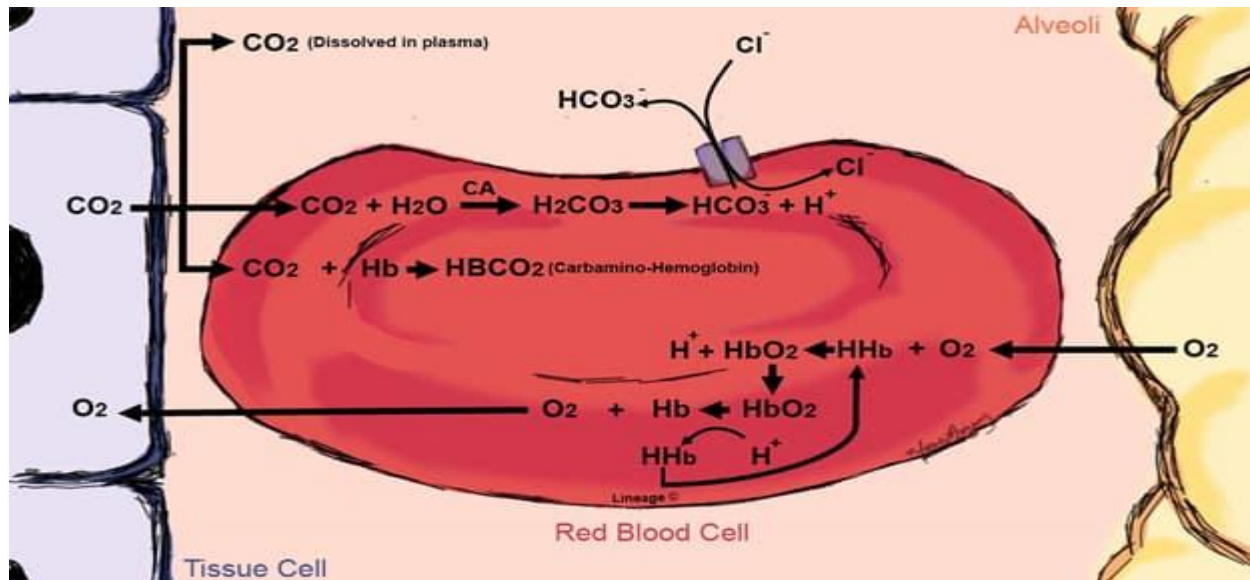
- They are **not true cells**, as they have **no nuclei, or organelles**.
- During **differentiation and maturation**, erythrocytes synthesize large amount of Hb, before they are released into the systemic circulation, the nucleus is extruded from the cytoplasm and the mature RBCs assumes a biconcave shape.
- The cytoplasm contains mainly **hemoglobin** (a cell inclusion) enclosed by cell membrane.
- Hemoglobin = the iron ("heme"), oxygen transport protein, ("globin").
- Biconcave shape provides more surface area for carrying respiratory gases.

- **The plasmalemma of the erythrocyte** consists of about 40% lipid, 10% carbohydrate, and 50% protein. The major components of the erythrocyte plasmalemma proteins are **Integral membrane proteins**, including **glycophorin A,B,C** and **band 3 protein**.
- **Glycophorin A** has antigenic sites that form the basis for the ABO blood system.
- **Several peripheral proteins** are associated with the inner surface of the membrane, including **spectrin** form a lattice bound to underlying actin filaments, and **ankyrin**, which anchors the lattice to the glycophorins and band 3 proteins.
- The erythrocyte cell membrane *and* the submembranous meshwork **maintain the cell shape, and provides the cell elasticity required for passage through capillaries.**



□ Functions of RBC:

- Mature RBCs are highly specialized to transport O₂ & Co₂.
- Transport of oxygen from lung to tissues and carbon dioxide from tissues to lung.
- Hb bind with O₂ and is carried to tissues in the form of **oxyhemoglobin**.
- Co₂ from the cells and tissues is carried to the blood with Hb in the form of **carbaminohemoglobin**. These reactions are reversible.



Abnormalities:

❖ In number of RBCs:

- **Anemia** is the condition of having a concentration of erythrocytes below the normal range.
- **Erythrocytosis, or polycythemia** An increased concentration of erythrocytes in blood, may be a physiologic adaptation found, for example, in individuals who live at high altitudes, where O₂ tension is low.

❖ In shape and size of RBCs:

- ▶ **Macrocytic anemia**, diameter of RBCs increases.
- ▶ **Microcytic anemia**, their diameter decreases.
- ▶ **Anisocytosis**, abnormal sizes of RBCs may appear in the circulation.

❖ In HB content of RBCs:

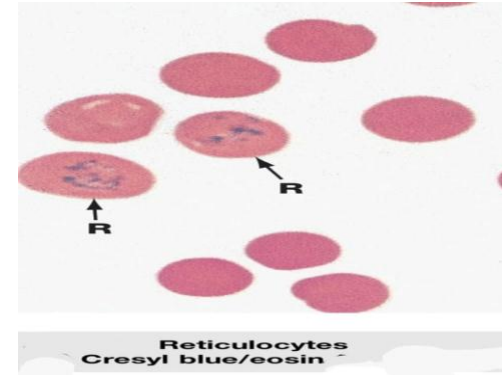
In **hyperchromic anemia**, the RBCs are darkly stained and contain more Hb % than normal.

- In **hypochromic anemia**, RBCs are faintly (lightly) stained and contain less Hb % than normal.

Reticulocytes

- They are **immature** (younger) erythrocytes.
- **Percentage:** constitute about **1%** of the total number of circulating RBCs.
- **Diameter:** is 8 μm . slightly larger than mature erythrocyte.
- **Structure:**

- ✓ having **no nuclei**
- ✓ **Cytoplasm** : Contains:
 - some **free ribosomes**
 - few mitochondria
 - Golgi elements
 - haemoglobin.

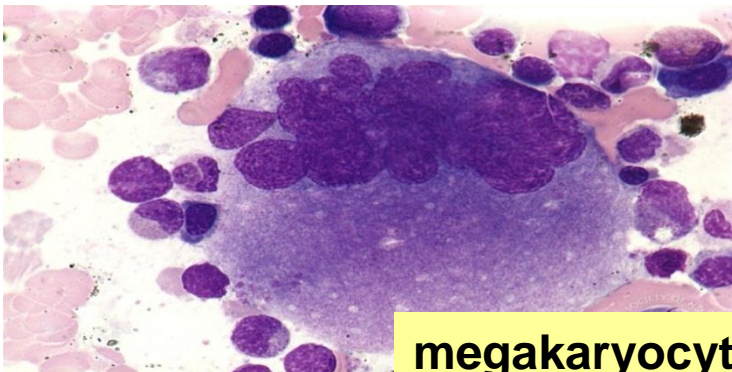


- **Stain:**
 - is slightly **basophilic** due to the ribosomes .
 - by **supravital stains** (**brilliant cresyl blue**) which stain **ribosomal RNA** that appear as blue stained network (reticulum) or few blue dots within the cell.
- **Abnormalities:** **reticulocytosis**

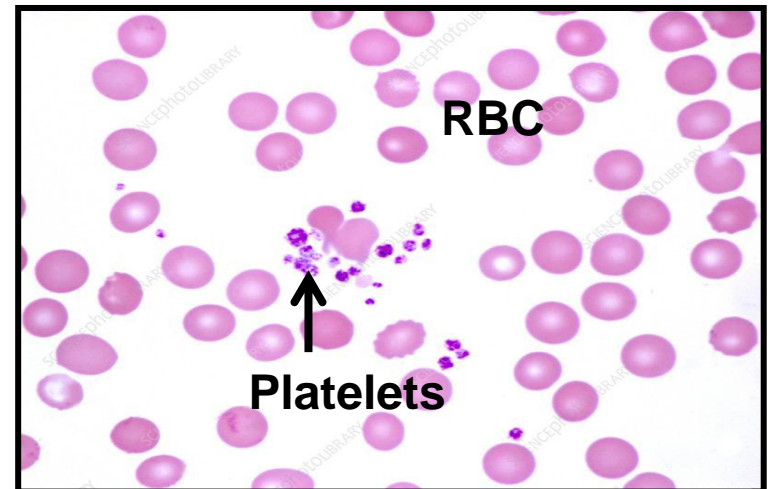
Increase the number of reticulocytes indicates a demand for increased O₂-carrying capacity as in *haemorrhage* and in *high altitude*.

Platelets (Thrombocytes)

- **Origin:** They are non nucleated cytoplasmic fragments derived from giant cells in the bone marrow called **megakaryocytes**.
- **Shape:** cytoplasmic fragments
- **Size:** 2-4 micron
- **Count:** 150,000 – 400,000 / **cubic millimeter** of blood.
- **Life span:** 5-10 days.



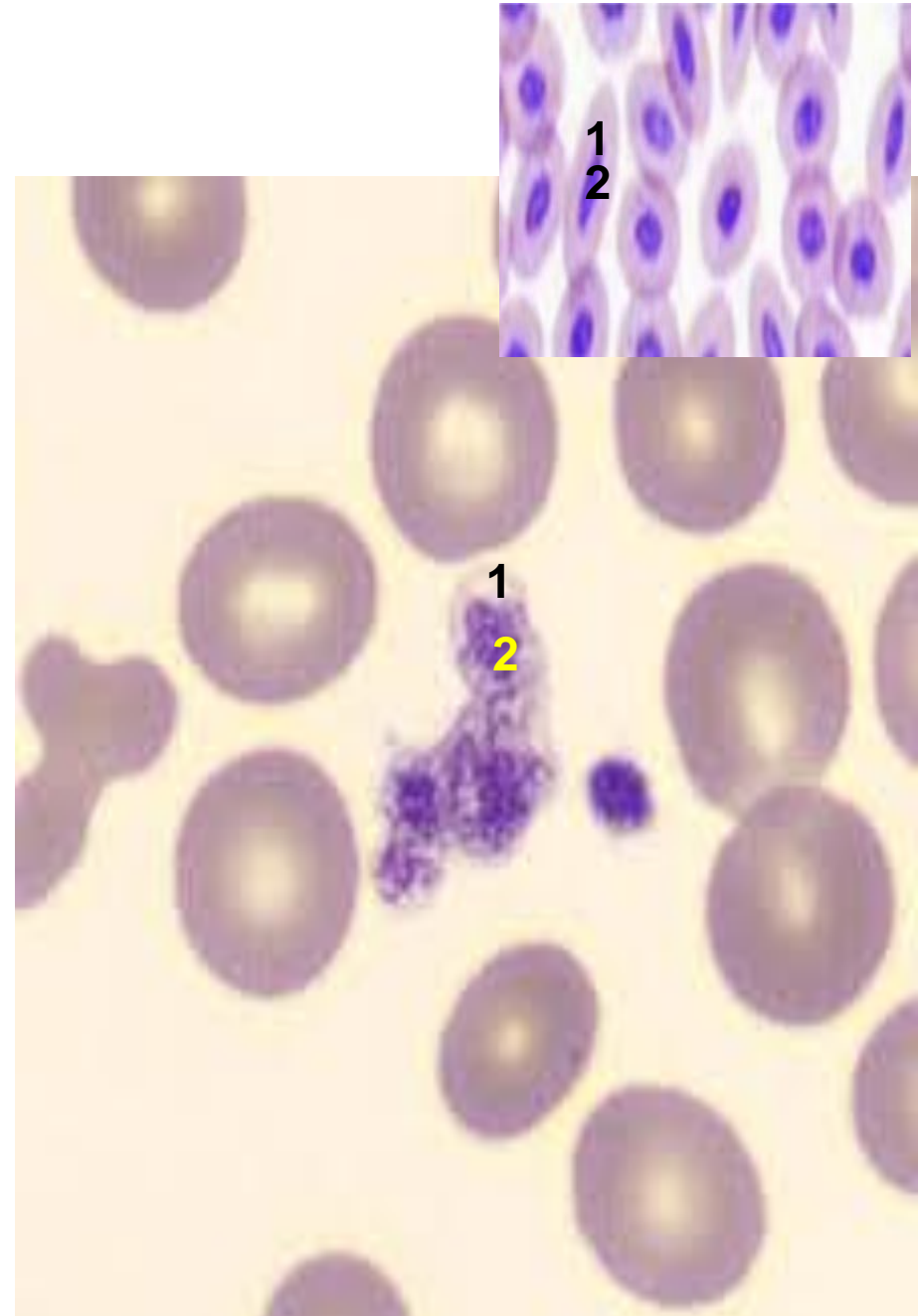
megakaryocytes.



- Structure:

- L/M:

- in stained blood smears, they often appear in clumps.
- *Each platelet:*
 - ✓ is formed of ovoid non-nucleated mass of cytoplasm surrounded by a cell membrane.
 - ✓ has a **peripheral** light blue-stained transparent zone called **hyalomere (1)**.
 - ✓ and a **central zone** containing purple granules called **granulomere (2)**.



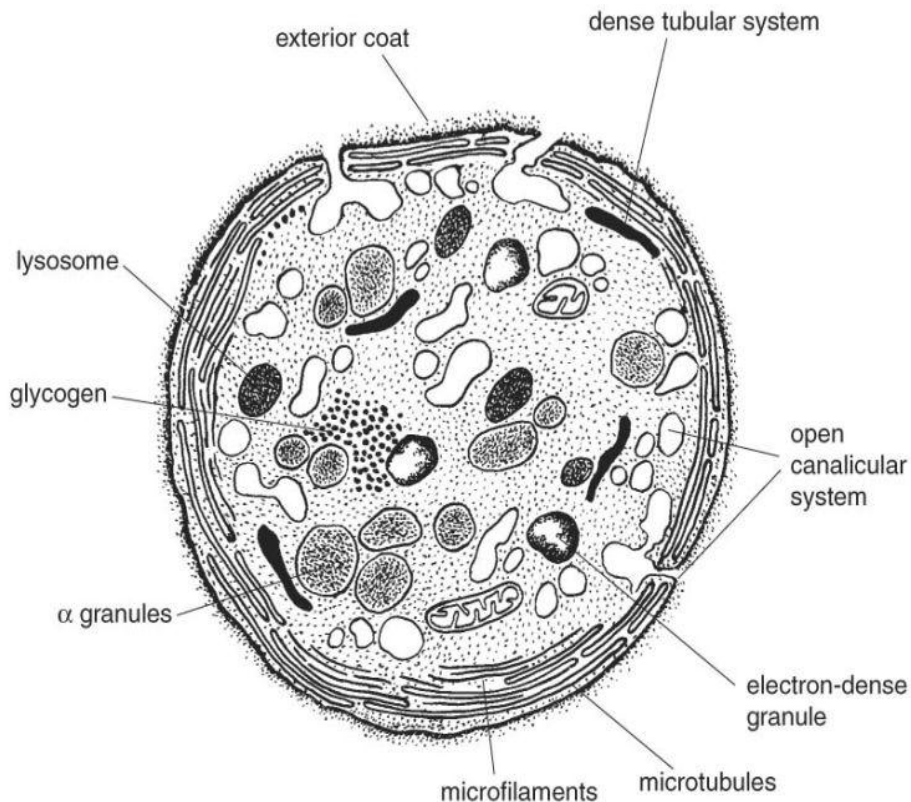
E/M: platelets are surrounded by cell membrane, covered by a thick glycocalyx (cell coat).

➤ The **hyalomere** contains 4:

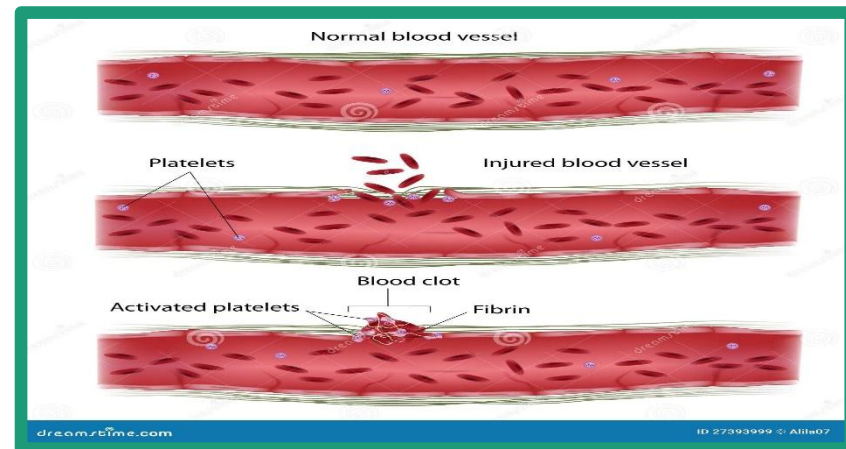
1. An open canalicular system of vesicles connected to invaginations of the plasma membrane for facilitating the liberation of the stored contents of the platelets.
2. A dense tubular system: Electron dense irregular tubules derived from the ER.
3. A bundle of microtubules which are arranged parallel to the cell membrane. They act as internal skeleton that maintains the oval shape of platelets.
4. Actin and myosin filaments (help in platelet movement and aggregation).

➤ The **granulomere** contains 4:

1. *Mitochondria and peroxisomes.*
2. *Glycogen granules.*
3. *Numerous vesicles which are invaginations of the cell membrane and cell coat.*
4. **Membrane-bounded dense granules named:**
 - **Alpha granules** contain fibrinogen and platelet specific proteins.
 - **Delta granules** contain calcium ions, ADP, ATP, and serotonin.
 - **Lambda granules** containing lysosomal enzymes.



Function: Promote blood clotting, so preventing loss of blood.



Leukocytes (white blood cells or WBCs)

- Leukocytes are **true cells** with a nucleus and cytoplasm.
- They are called leukocytes because they are colourless (No Hb).

❑ Total leukocytic count:

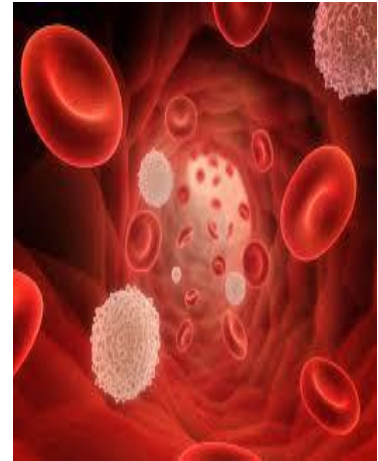
4,000 – 11,000 / cubic millimeter of blood.

❑ Origin :

- **All WBCs** are formed in the **bone marrow** in postnatal life.
- All WBCs mature in bone marrow **except T-lymphocytes** mature in **thymus**.
- They originate in the bone marrow, circulate in the blood and then migrate to the loose connective tissue, where they perform their functions that related to immunity.

❑ Life span:

- Life span of leucocytes ranges from **hours to one day** in the circulation.
- So, to maintain normal circulating blood leucocytes so body produces over 100 billion cells per day.
- Normally **75% of cells** in the bone marrow belong to WBCs and only **25% are maturing red cells**.



Classification of Leukocytes

leukocytes are classified according to the presence or absence of **specific granules** (contain substances unique to each cell's function) in their cytoplasm when stained into:

I- Granular leukocytes:

- Their cytoplasm contains **two types** of granules :

1- **Specific** granules

2- **Non-specific** granules

- They are further **subdivided into 3 types** according to the staining properties of their specific granules :

1. **Neutrophils.**

2. **Eosinophils.**

3. **Basophils**

II- Non-granular leukocytes:

- Their cytoplasm contains **only non-specific** granules.
- They are further subdivided into:

1. **Lymphocytes.**

2. **Monocytes.**

The contents of these granules (**cytotoxic substances** and/or **active inflammatory mediators**) play a unique role in immune response

Classification

Granular Leukocytes

Neutrophils
60-70%

Eosinophis
1-4%

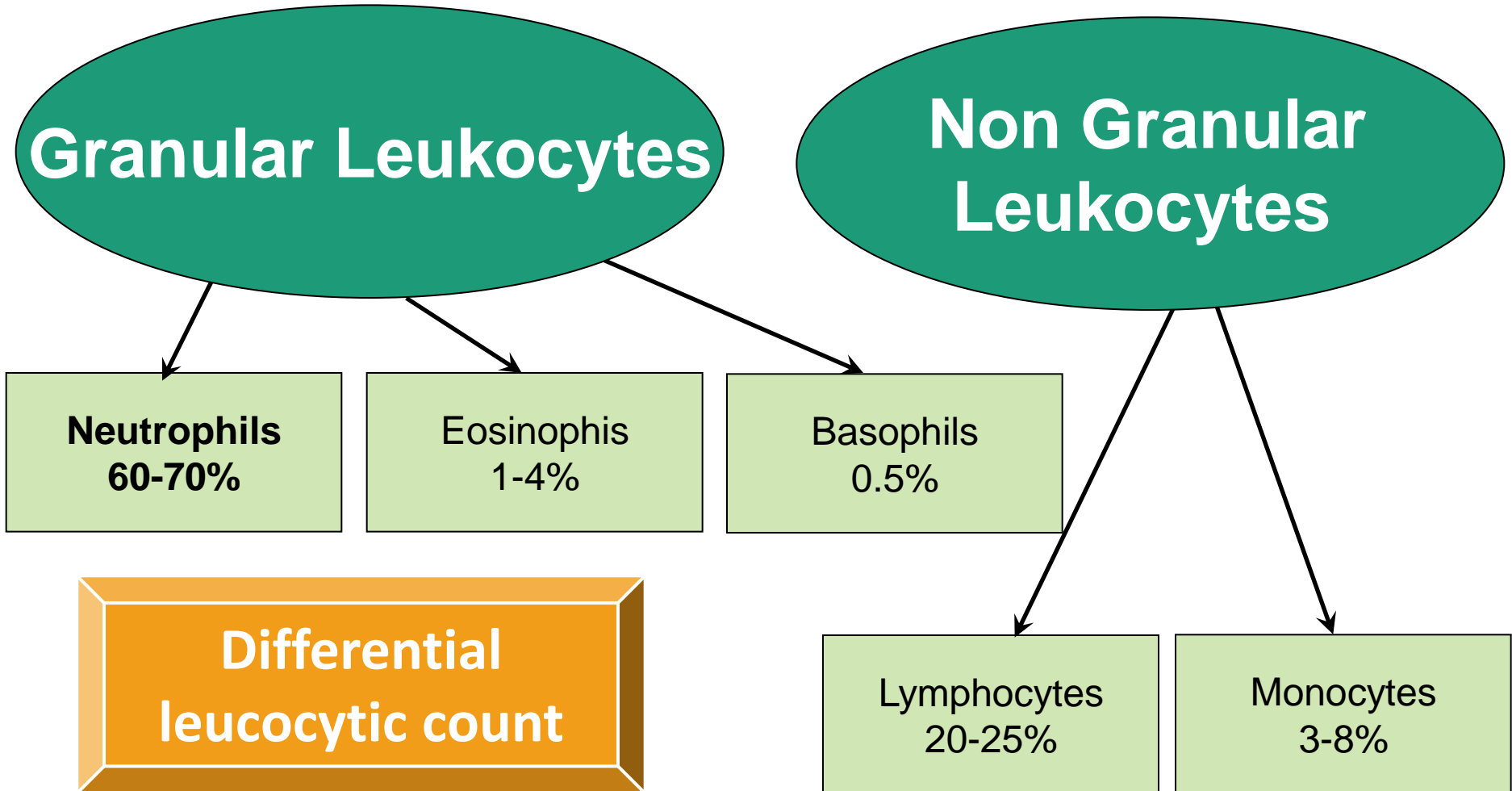
Basophils
0.5%

Non Granular Leukocytes

Lymphocytes
20-25%

Monocytes
3-8%

**Differential
leucocytic count**



Leukocytosis is the increase in number of leukocytes above 11000 / cubic millimeter which is either:

- **Physiological:** as during pregnancy, lactation, after muscular exercise.
- **Pathological:** as in acute pyogenic infections (abscess, acute tonsillitis and acute appendicitis).

Leucopenia is the decrease in the number of WBCs below 4000/ cubic millimeter, it occurs in:

- Influenza.
- Exposure to irradiation & X-ray.

Common properties of leukocytes:

1- Margination:

attracted to the capillary endothelial surface and **roll** along it.

2- Diapedesis:

can **squeeze** through narrow pores in the walls of capillaries.

3- Motility:

have amoeboid movement by develop pseudopodia.

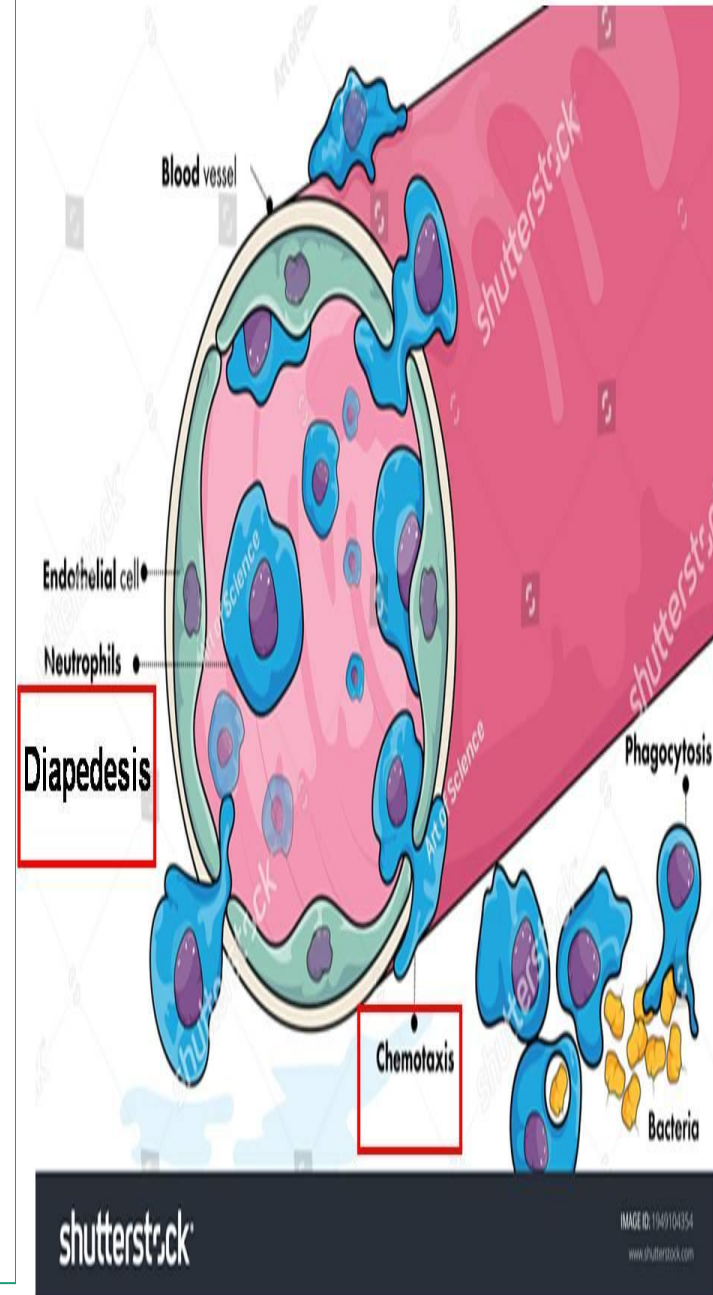
4- Chemotaxis:

Certain substances attract the leucocytes.

- **Examples:** degenerative products of inflamed tissues and some bacterial toxins.

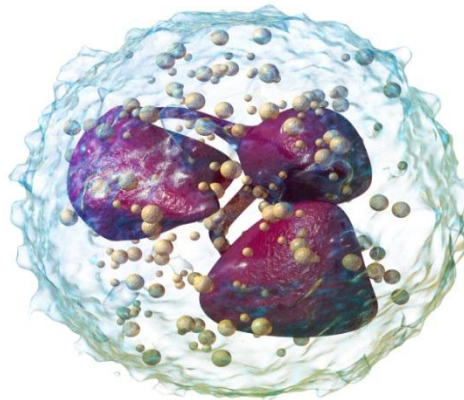
5- Phagocytosis:

It means that the cells can **ingest** the microbes



Neutrophils (Polymorphnuclear leukocytes)

- They are **circulating** in blood and could migrate to the inflammatory tissues.
- Neutrophils are the **most abundant** population of the **circulating** WBCs (50-70%)
- *They have properties* of margination, diapedesis, motility, chemotaxis and phagocytosis of bacteria.



- **Structure:**

LM:

Shape: rounded cells

The nucleus: lobulated, darkly stained.

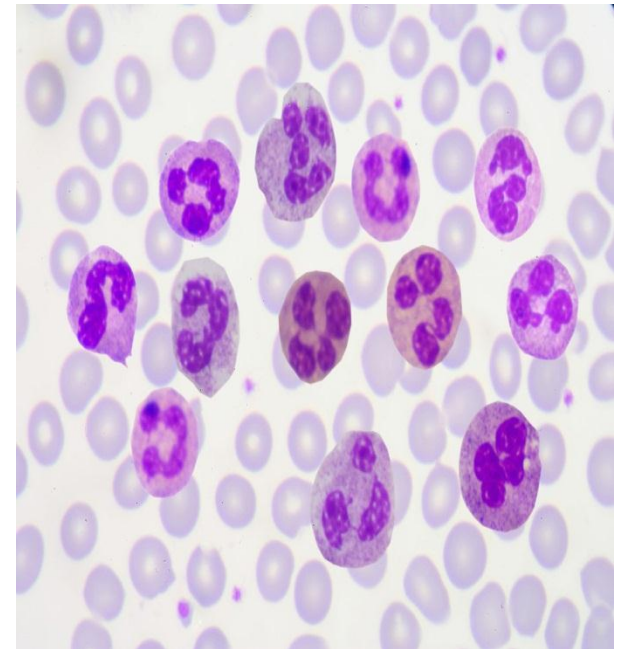
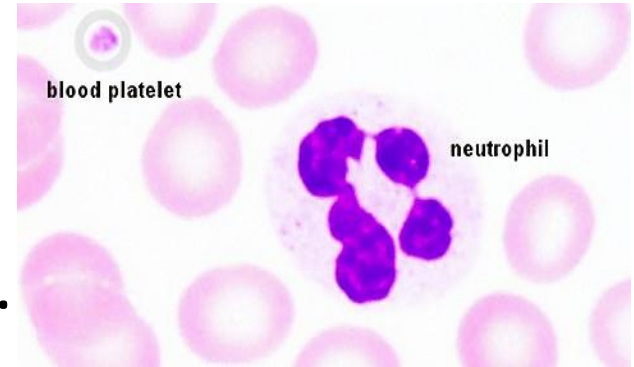
EM:

The nucleus

- ✓ **Segmented** into many lobes (usually 3) connected to each other by chromatin threads.
- ✓ Contains *condensed chromatin*.

Due to the variability of the nuclear shape, the neutrophils are usually called

polymorphonuclear leukocytes



Cytoplasm:

Surface:

- Have *Pseudopodia*
- Contains two types of granules:

✓ Specific granules

-Small & numerous (80%)

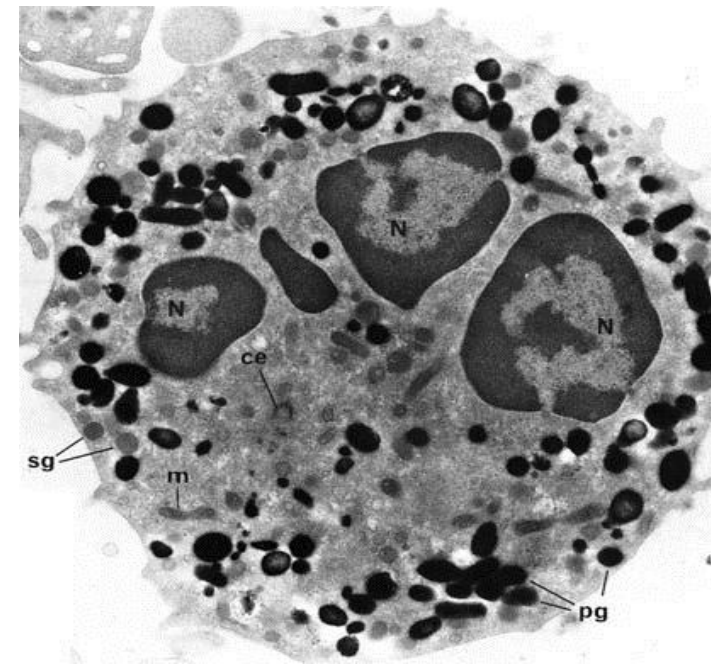
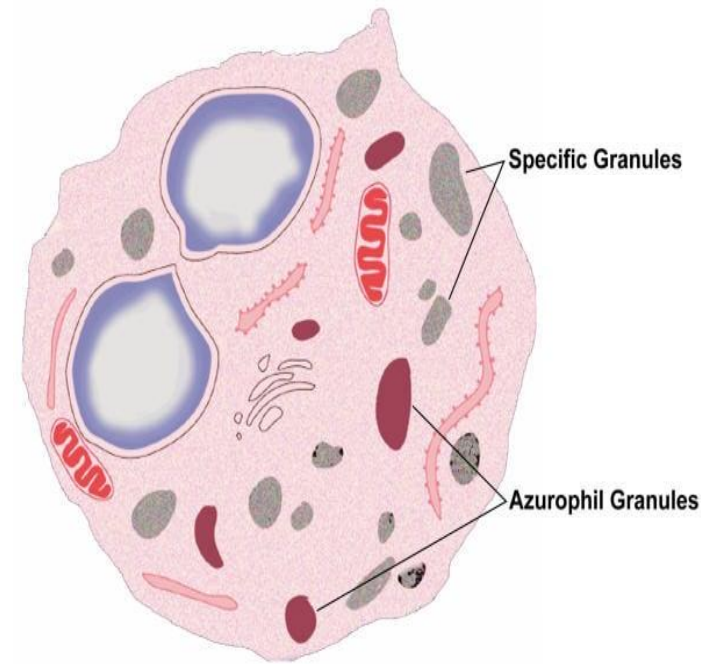
-EM: These granules are **membranous vesicles** containing **ECM-degrading enzymes such as collagenases and bactericidal enzymes.**

✓ Non-specific (Azurophilic) granules

-Large, less numerous(20%)

-These granules are **primary lysosomes** containing **hydrolytic enzymes.**

✓ The cytoplasm also contains glycogen, small Golgi body, few mitochondria and little endoplasmic reticulum.



- **Function of neutrophils:**

- ✓ The main function is **Phagocytosis** & digestion of micro-organisms especially **bacteria** by **the specific and non specific granules** (They exert a powerful defense action mainly against **bacterial infections**).
- ✓ The **first phagocytic** cells that normally encounter the foreign molecules and microbes in blood. So it is called **microphage**.
- ✓ They are important in **inflammatory response** as it releases **chemotactic factors** that stimulate attraction of other immune cells to the site of inflammation.

- **Abnormal count:**

Neutrophilia means increase in the percentage of neutrophils above normal (as in acute bacterial infections).

-**Neutropenia** means decrease in the percentage of neutrophils below normal (as in viral infections).

Eosinophils

- ❑ 1-4% of total leukocytes.
- ❑ They are abundant in the gastrointestinal and respiratory tracts mucosa.

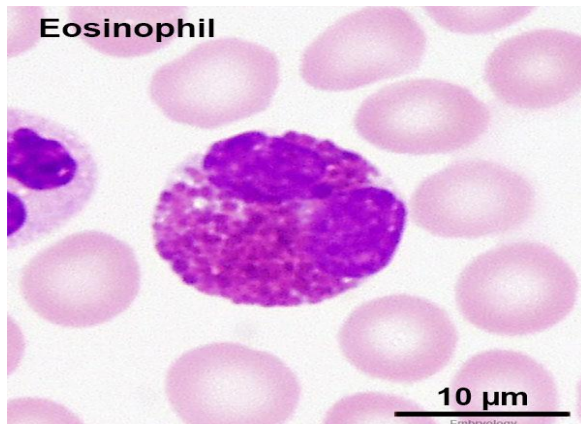
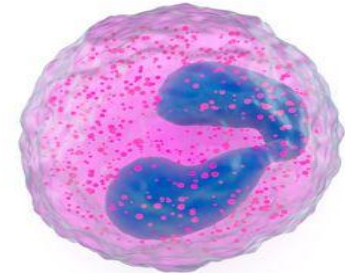
❑ Structure:

LM:

- The cell is **rounded**.
- Containing **large acidophilic** cytoplasmic granules.

The nucleus:

- **Bilobed** and connected by a thin chromatin giving it **horse-shoe shaped** appearance
- Paler than that of neutrophil.



EM:

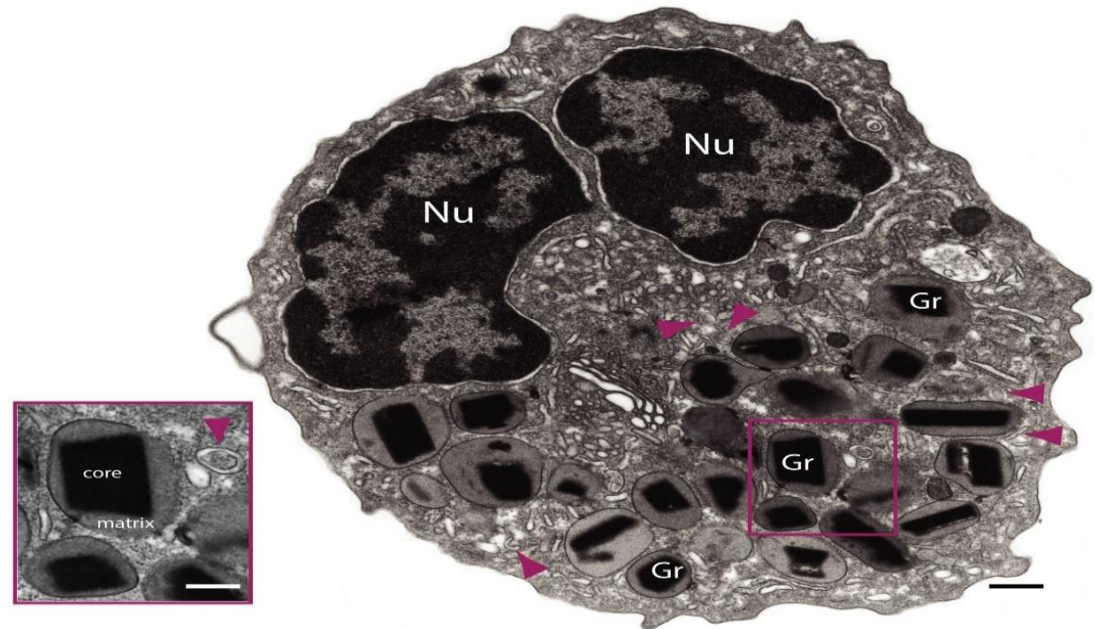
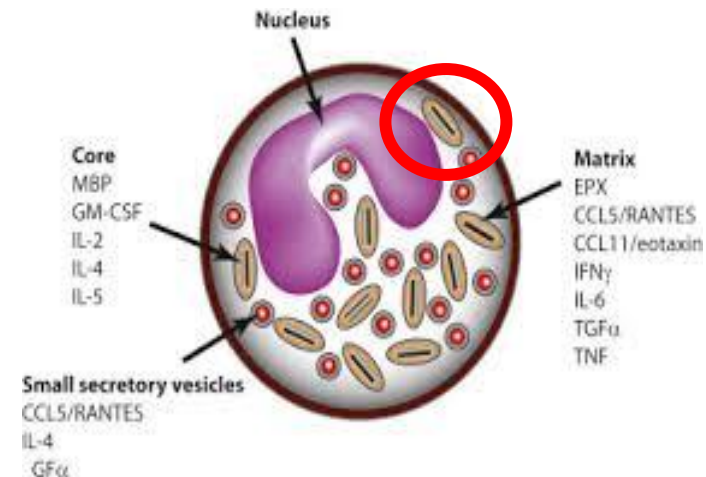
Cytoplasm: contains:

✓ Specific granules:

- **Large elongated specific granules** with central crystalline dense core formed of protein called **major basic protein (MBP)**.
- This core is surrounded by less dense material consists of some enzymes as **histaminase**.

✓ Non specific granules are **lysosomes** containing hydrolytic enzymes.

✓ Glycogen, and poorly developed endoplasmic reticulum, mitochondria and Golgi body are present.



- **Function:**

- ✓ They are **attracted to the sites of allergic reactions** by *eosinophil chemotactic factor* which is released by mast cells to reduce their effects by releasing antihistamine (histaminase).
- ✓ Play a role in *killing parasitic worms* by *major basic protein*

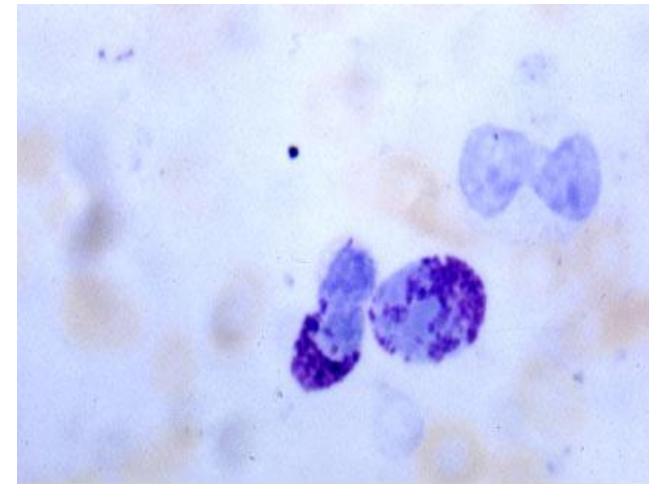
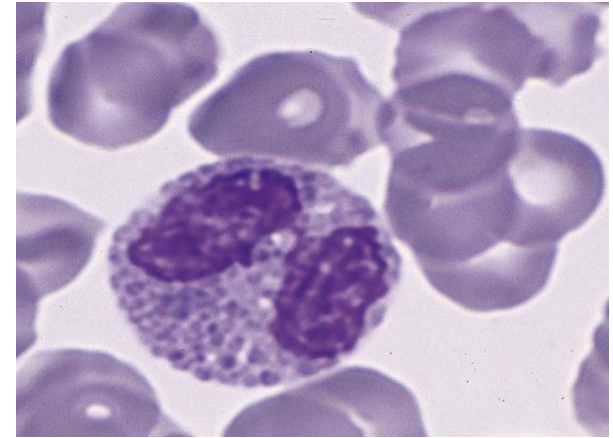
- **Abnormal count:**

Eosinophilia: in allergic and parasitic diseases.

Eosinopenia: after cortisone treatment.

Basophils

- ❑ 0.5% of total leukocytes.
- ❑ **Shape:** rounded
- ❑ **Structure:**
 - ❑ **Nucleus:** large -is often bent into a U or S shaped.
 - ❑ **Cytoplasm :**
 - ✓ Specific granules: they are large, basophilic and obscure the nucleus.
 - They are metachromatically stained by toluidine blue
 - contain heparin & histamine, like mast cells.
 - ✓ Non-specific granules: they are lysosomes

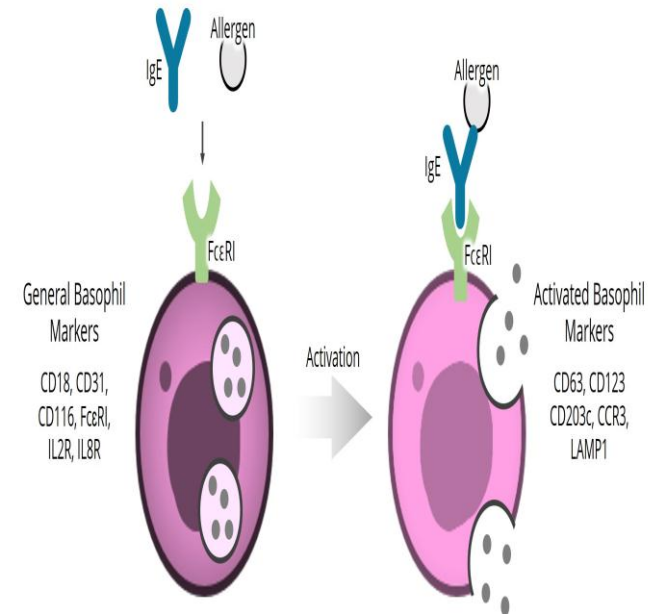


- ✓ Contains varying amounts of glycogen, small Golgi apparatus, a few mitochondria and poorly – developed endoplasmic reticulum.
- ✓ The cell surface has receptors for **the plasma IgE**, which when come in contact with the antigen (in the blood); they form antigen-antibody complex resulting in degranulation of the basophils and release of its mediators.



□ Functions:

- Secretion of **eosinophil chemotactic factor**.
- Secretion of **heparin** (anticoagulant).
- Secretion of **histamine** (initiates allergic reactions).

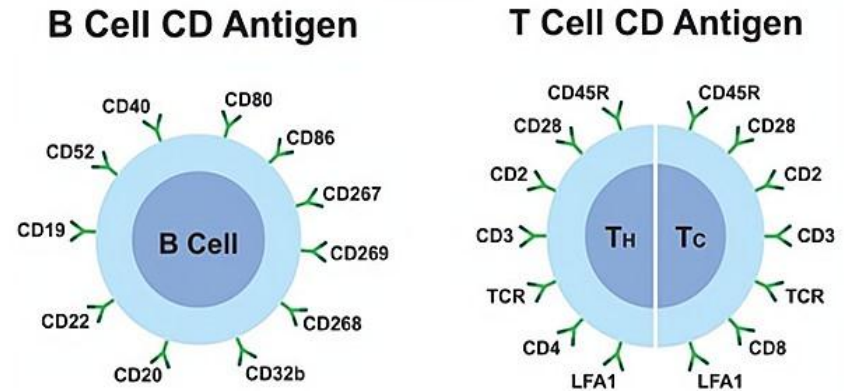


Lymphocytes

✓ 20-25% of total leukocytes.

✓ Lymphocytes can be subdivided into **functional groups** according to certain surface molecules expressed on the cells (cluster of differentiation or CD markers) into :

- **B lymphocytes**
- **T lymphocytes**
- **Natural killer (NK) cells.**



✓ Histologically, they can be classified according to their size into:

- **Large lymphocytes.**
- **Small lymphocytes.**

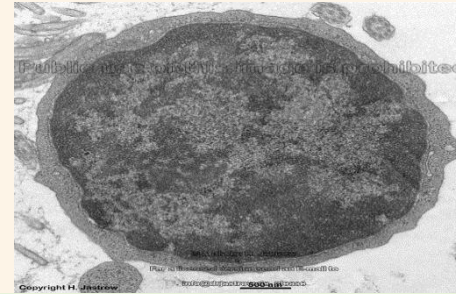
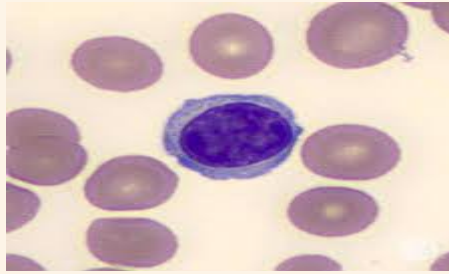
✓ They are present in the C.T., lymph nodes, spleen, thymus, tonsils and tissue fluids.

✓ **Small lymphocytes:** They are the commonest. **Types:** T- lymphocytes , B-lymphocytes

-**Shape:** spherical

-**Nucleus:** is large, rounded & darkly stained (condensed chromatin) with little indentation at one side.

-**Cytoplasm:** is scanty, and appears as a narrow rim around the nucleus. It is lightly basophilic and non-granular containing a few azurophilic granules, mitochondria, a small Golgi complex and abundant ribosomes.

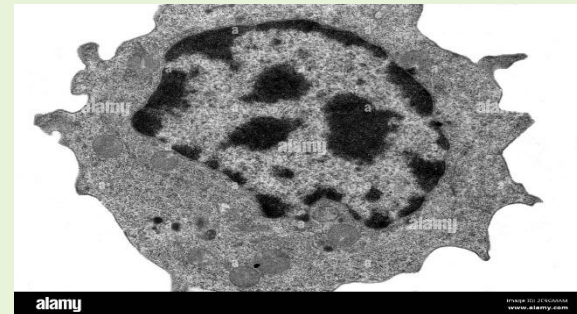
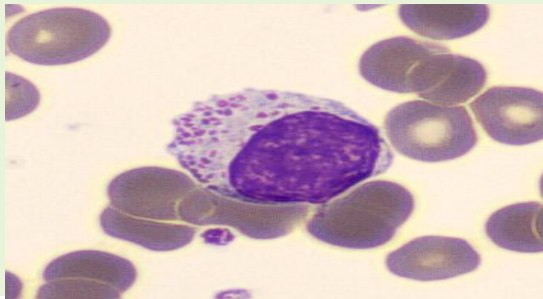


✓ **Large lymphocytes:**

They are believed to be small lymphocytes activated by the specific antigens.

-**Nucleus:** is large & lightly stained (active chromatin) with apparent nucleolus.

-**Cytoplasm:** is abundant, more basophilic (containing ribosomes) & non-granular. It also contains a few azurophilic granules.



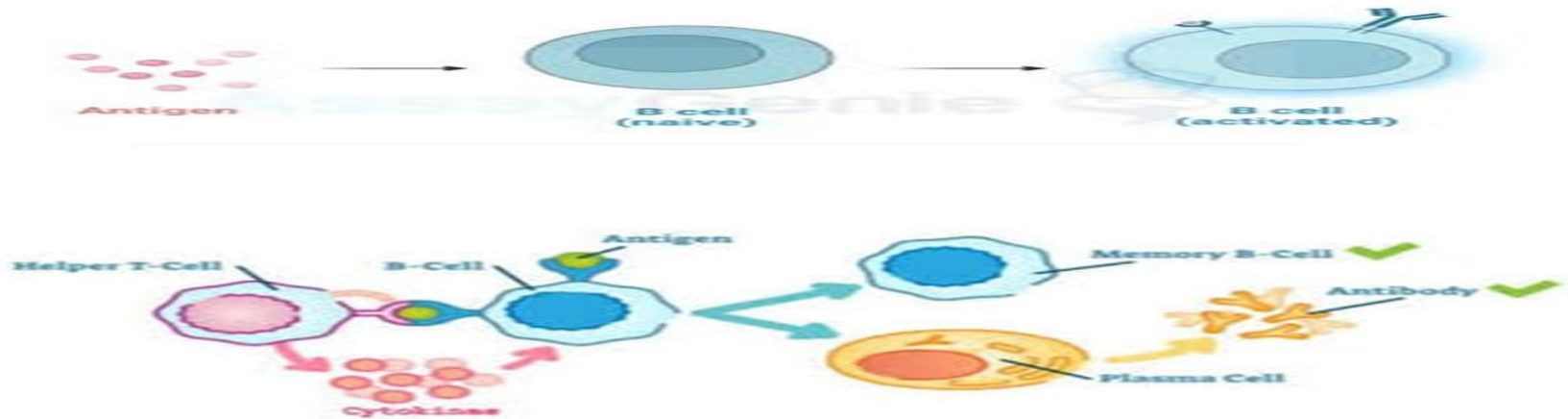
Function of lymphocytes:

✓ **T-lymphocytes:** They are responsible for cellular immunity.

✓ **B-lymphocytes**

- They are responsible for **humoral immunity**.

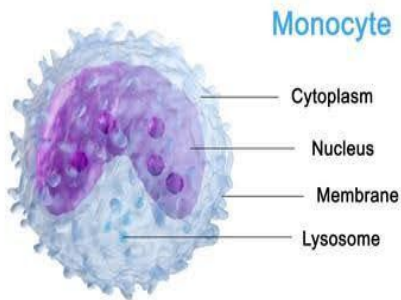
- when stimulated by specific antigen, some of **activated B-lymphocytes** differentiate into **plasma cells** to produce **antibodies**. Others generate **B-memory cells**, which react rapidly to a second exposure to the same antigen



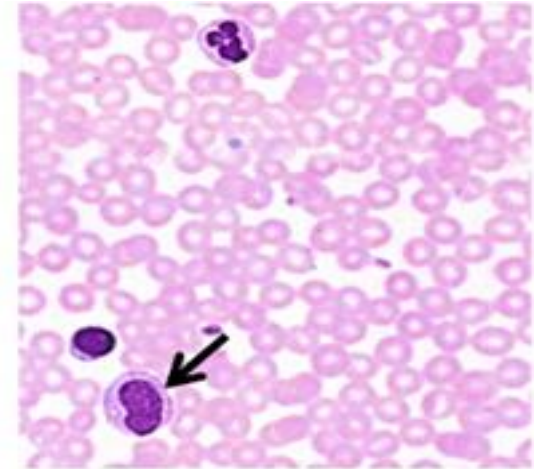
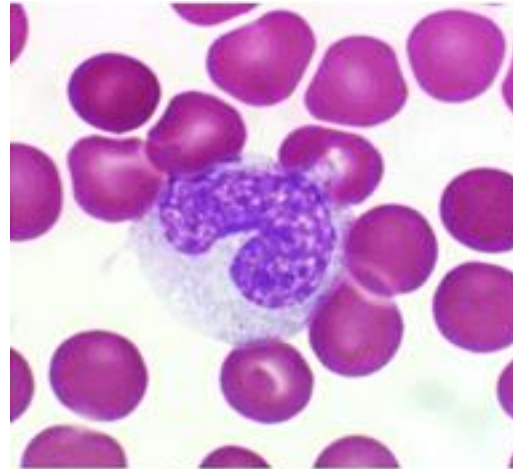
Activation of B-lymphocytes

Monocytes

- 3-8% of total leukocytes.
- Monocytes circulate in blood about **three days** after which they leave blood to the connective tissues, where they differentiate into **macrophages**.
- **Structure:** rounded cell



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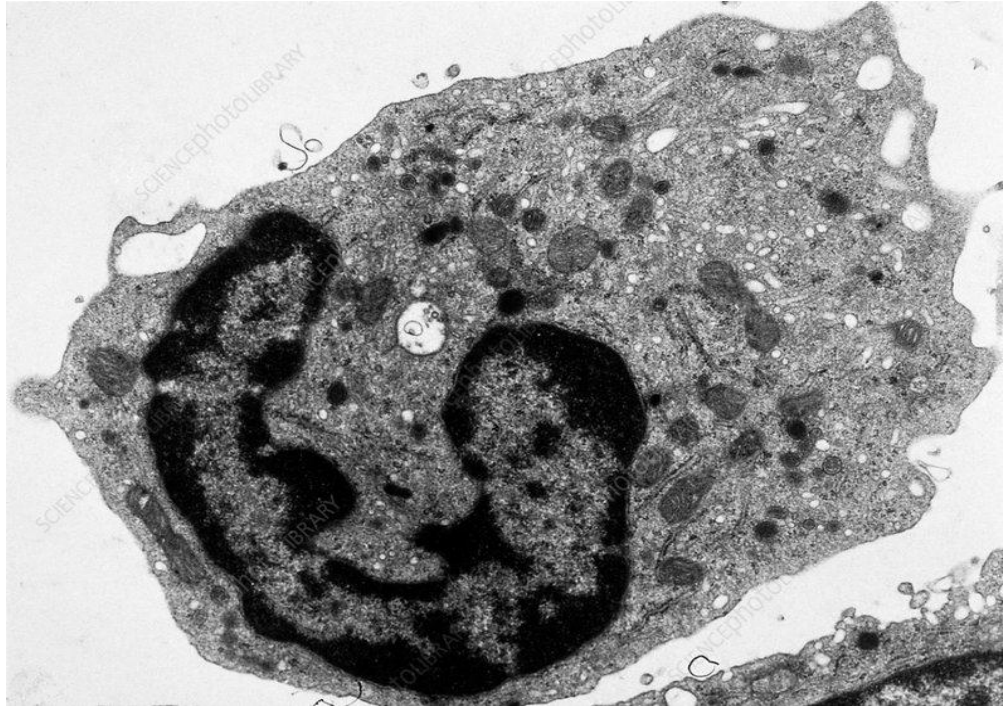


The nucleus:

- Oval in shape with deep indentation.
- Sometimes, it takes the **kidney shape**.
- Eccentrically placed.

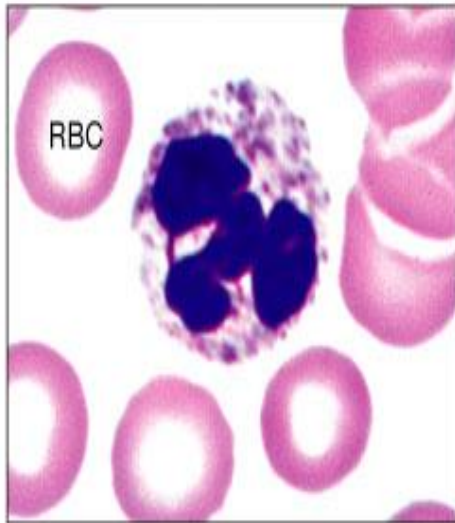
Cytoplasm:

- *The surface* shows pseudopodia.
- Abundant and pale blue.
- -Contains very fine **azurophilic granules (lysosomes)**, well developed Golgi.

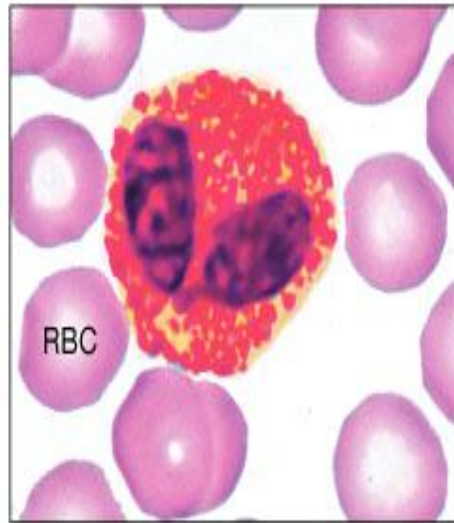


Function:

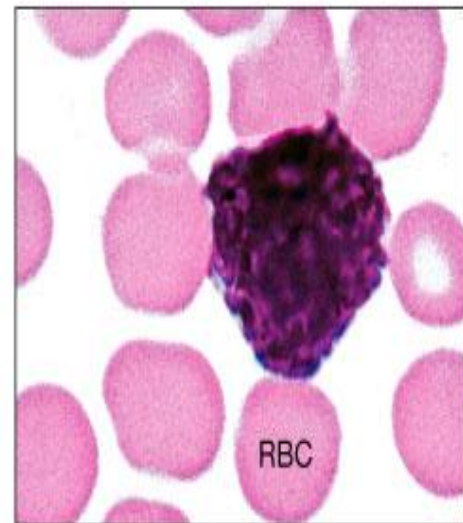
- In the connective tissue they change to macrophages which are **highly phagocytic cells**.
- Antigen presenting cell.



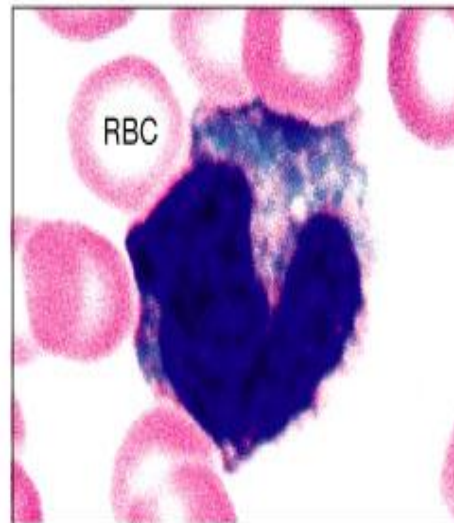
(a) Neutrophil



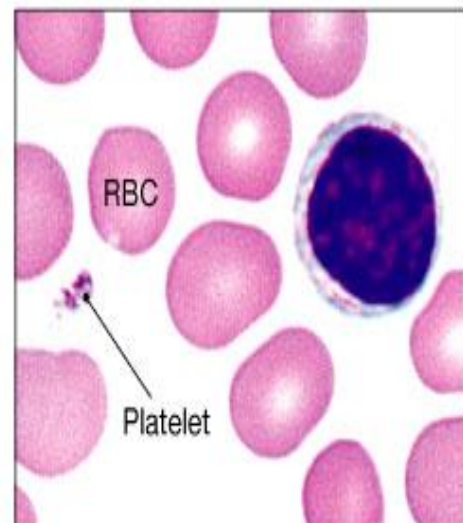
(b) Eosinophil



(c) Basophil



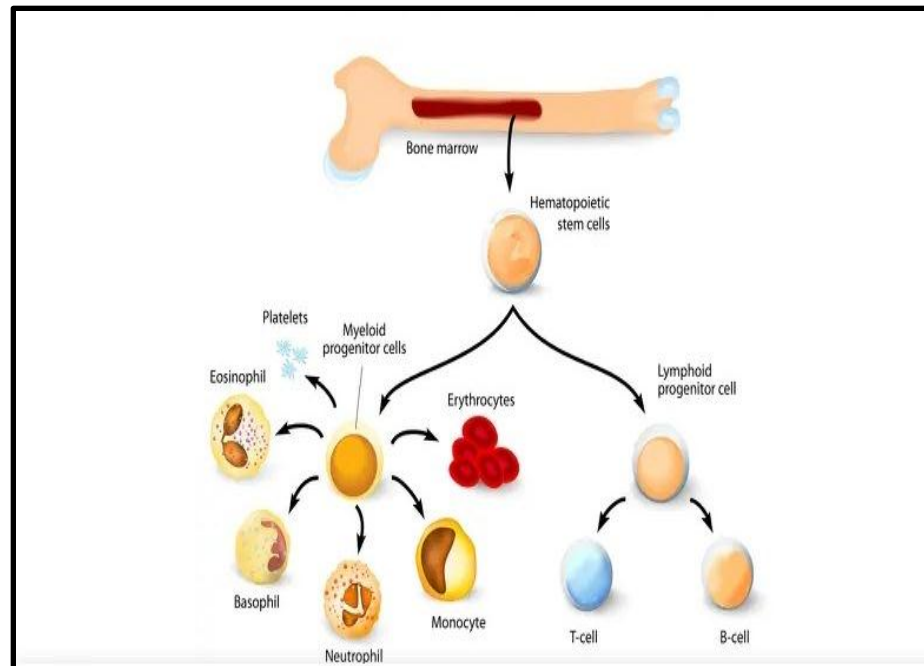
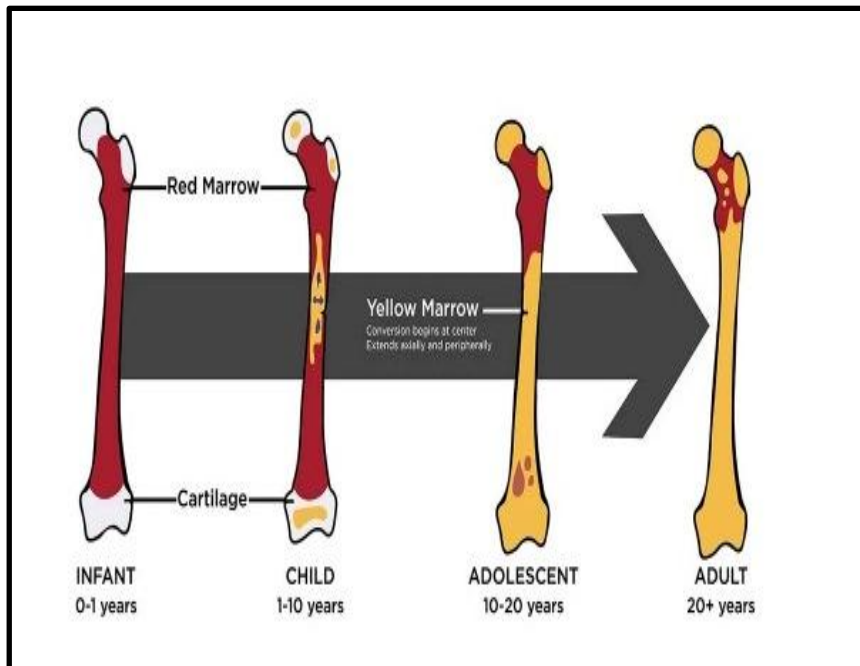
(d) Monocyte



(e) Lymphocyte

Bone marrow (Myeloid Tissue)

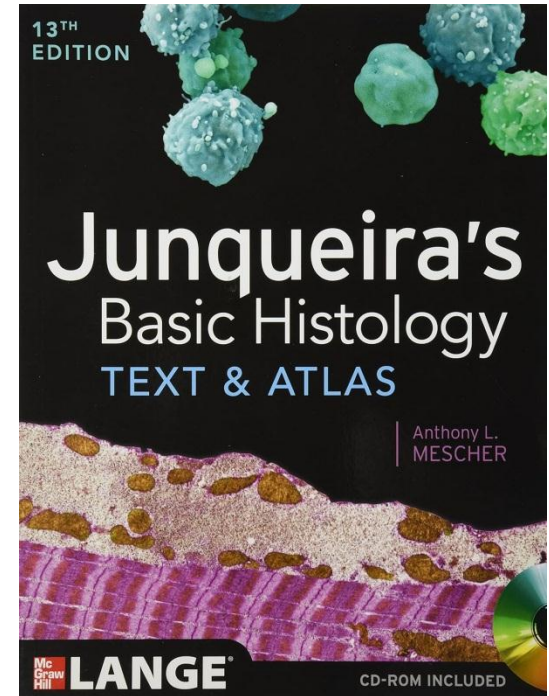
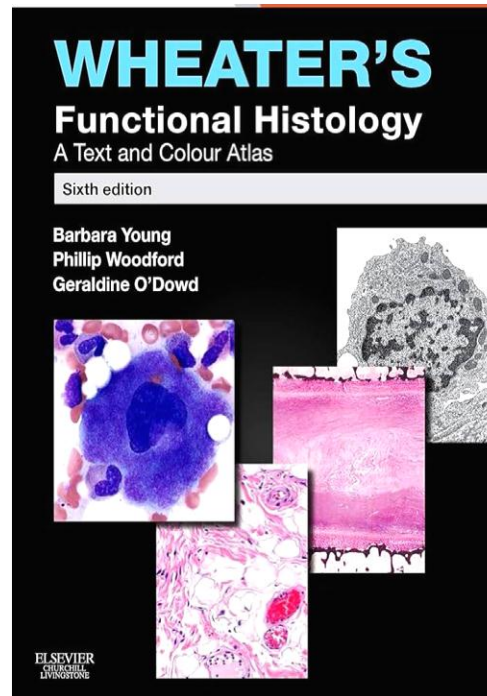
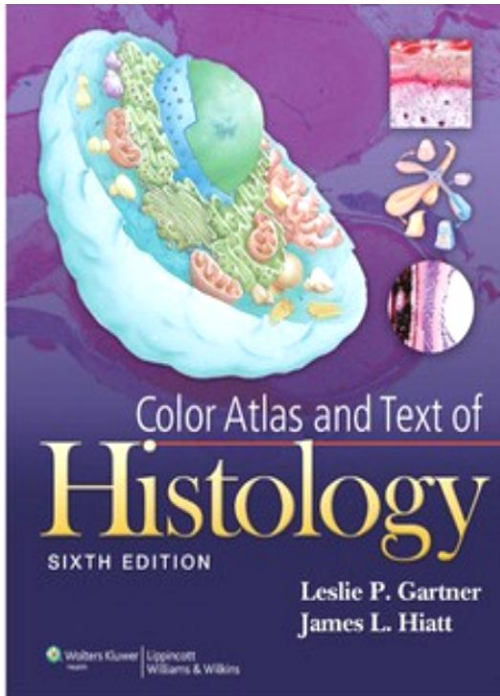
- The myeloid tissue is a **specialized vascular connective tissue** rich in cells that are responsible for formation of blood cells.



Types of bone marrow

	Red	Yellow
Activity	Active	Inactive
Site	In fetus present in all bones. In adult present in spongy bone.	Adult long bones
structure	<ul style="list-style-type: none">• <u>Stroma:</u> A delicate network formed of reticular CT. containing developing blood cells (<u>Hematopoietic cords</u>) in its meshes.• <u>Sinusoidal capillaries</u>	Yellow in color due to great number of adipose(fat) cells.
function	Formation of blood cells	<ul style="list-style-type: none">• Stores fat.• <u>Under certain conditions,</u> such as severe hemorrhage, becomes active and forms blood cells.

References



A vibrant watercolor splash in shades of yellow, orange, red, purple, and blue serves as the background. The words "Thank you" are written in a white, cursive script with a subtle drop shadow, centered over the splash. The overall composition is artistic and celebratory.

Thank you