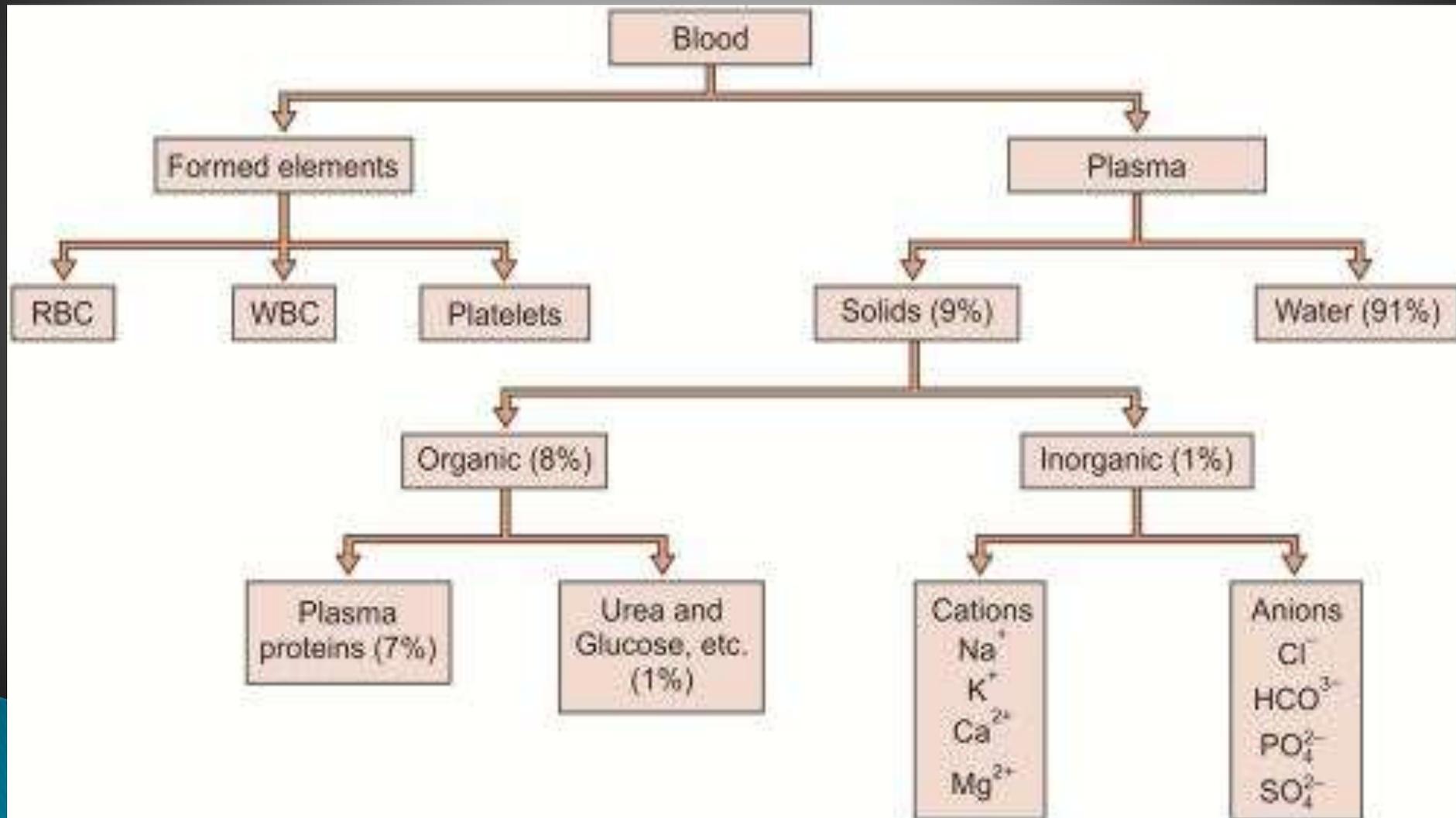


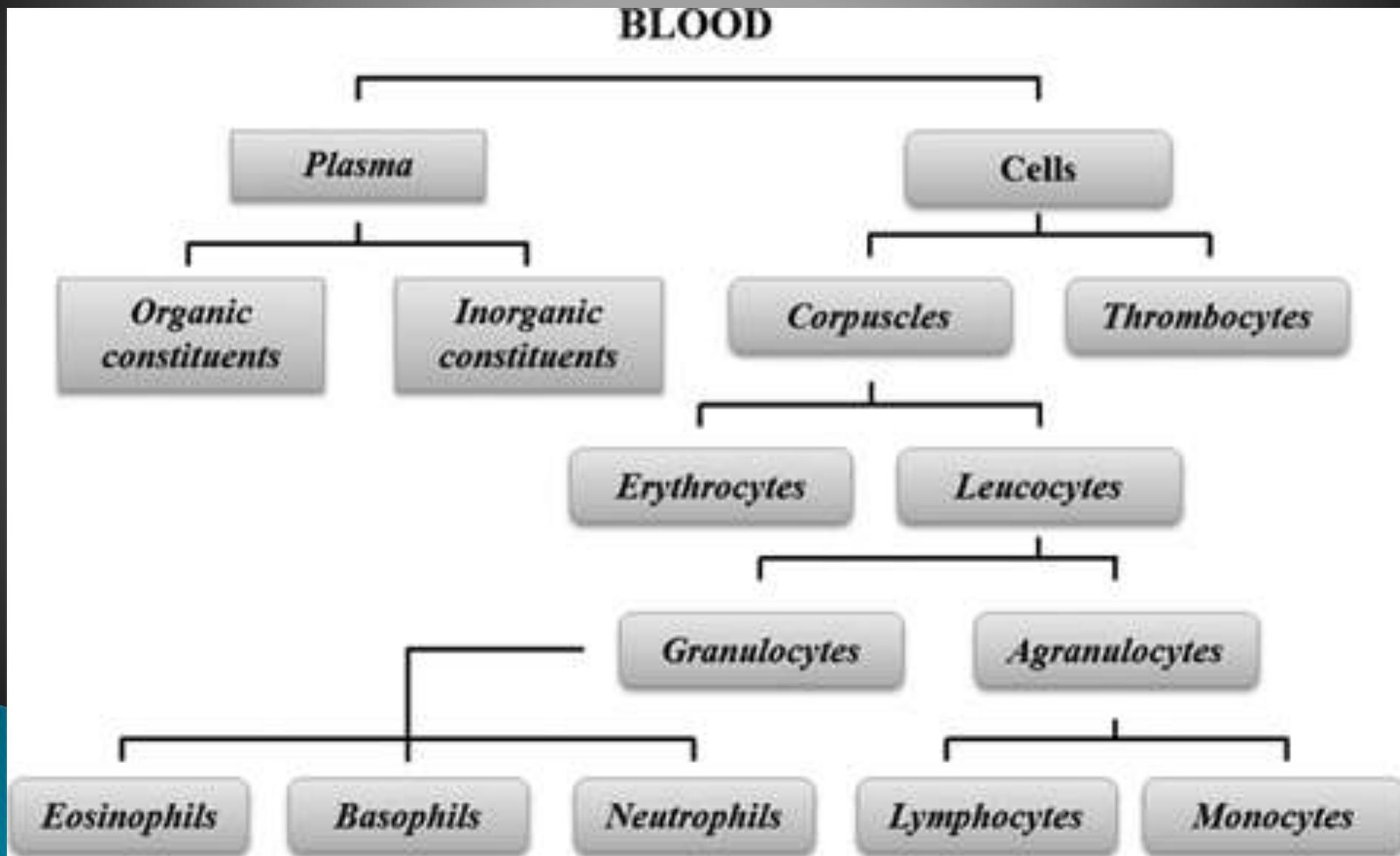
# ESTIMATION OF WHITE BLOOD CELLS

DR.SHEETAL JAIN

# BLOOD COMPOSITION

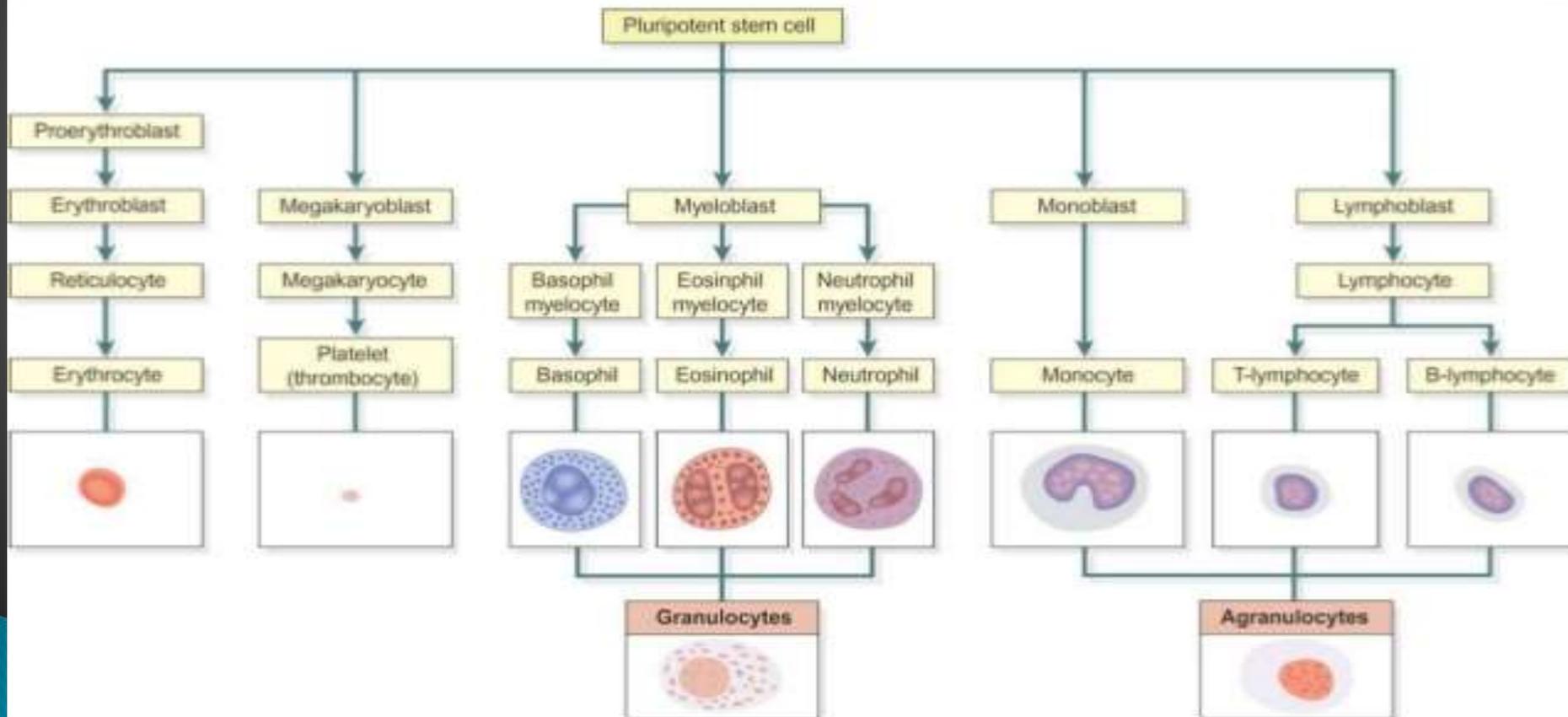


# BLOOD COMPOSITION

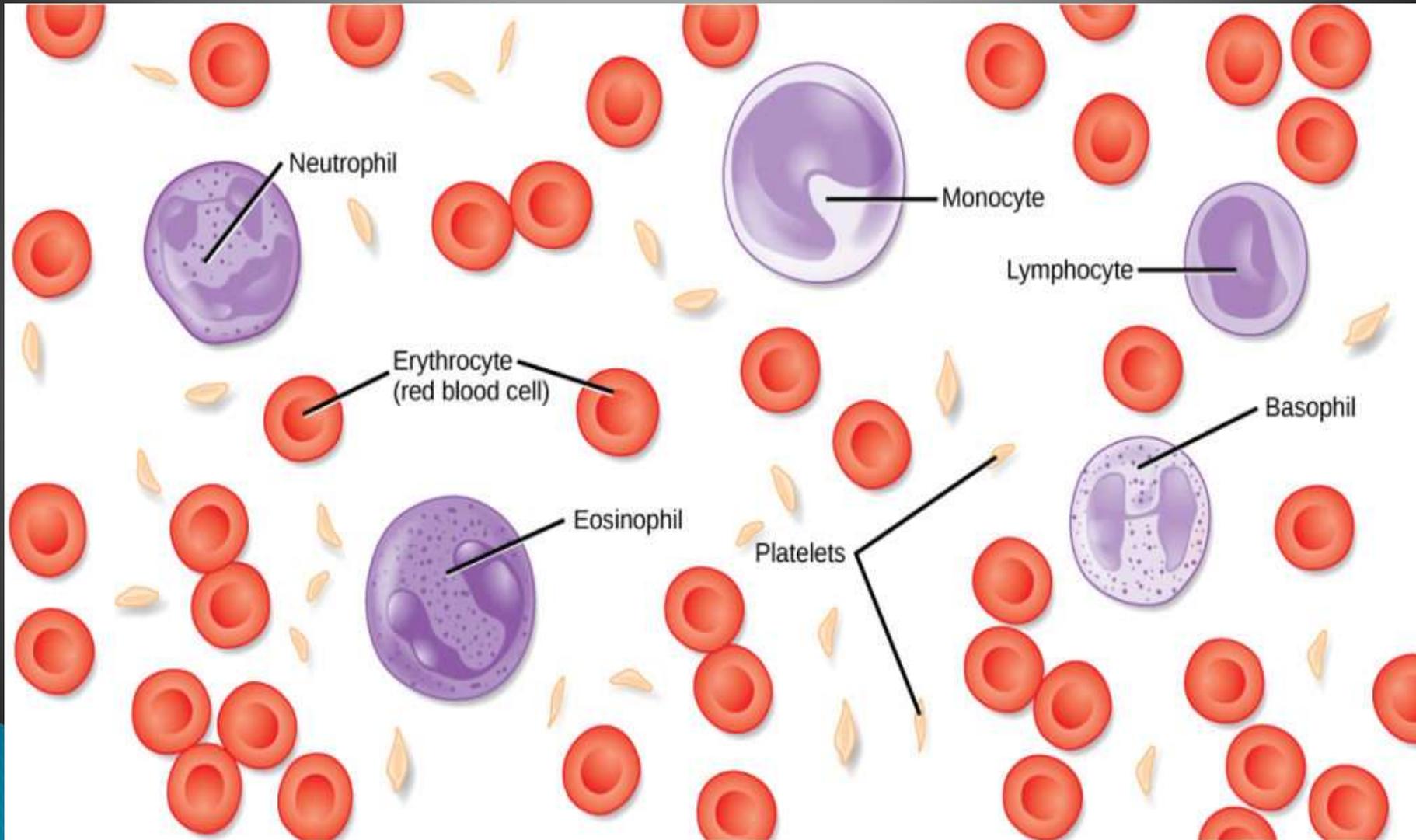


# BLOOD CELLS

## Haemopoiesis: Stages in the development of blood cells



# DIFFERENT TYPE OF BLOOD CELLS





Neutrophils



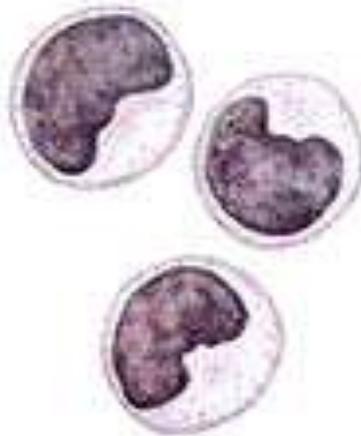
Eosinophils



Basophils



Lymphocytes



Monocytes

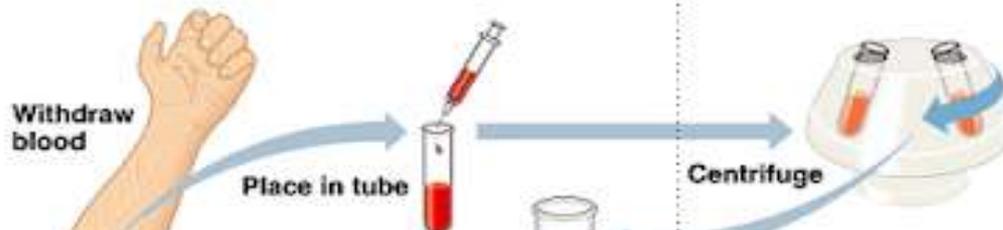


Platelets



Erythrocytes

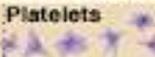
# Components of the Blood



## Plasma 55%

Constituent	Major functions
Water	Solvent for carrying other substances
Ions Sodium Potassium Calcium Magnesium Chloride Bicarbonate	Osmotic balance, pH buffering, and regulation of membrane permeability
Plasma proteins Albumin Fibrinogen Immunoglobulins (antibodies)	Osmotic balance pH buffering Clotting Defense
Substances transported by blood Nutrients (e.g., glucose, fatty acids, vitamins) Waste products of metabolism Respiratory gases (O <sub>2</sub> and CO <sub>2</sub> ) Hormones	

## Cellular elements 45%

Cell type	Number (per mm <sup>3</sup> of blood)	Functions
Erythrocytes (red blood cells) 	5–6 million	Transport oxygen and help transport carbon dioxide
Leukocytes (white blood cells)  Basophil  Eosinophil  Neutrophil  Lymphocyte  Monocyte	5000–10,000	Defense and immunity
Platelets 	250,000–400,000	Blood clotting

Type of WBC	Normal percentage of overall WBC count
neutrophil	55 to 73 percent
lymphocyte	20 to 40 percent
eosinophil	1 to 4 percent
monocyte	2 to 8 percent
basophil	0.5 to 1 percent

# AVERAGE WBC COUNT

Age range	WBC count (per mL of blood)
newborns	9,000 to 30,000
children under 2	6,200 to 17,000
children over 2 and adults	5,000 to 10,000

# LEUKOPENIA

Your blood is made up different types of blood cells, including white blood cells, or leukocytes. White blood cells are an important part of your immune system, helping your body to fight off diseases and infections. If you have too few white blood cells, you have a condition known as leukopenia.

There are several different types of leukopenia, depending on which type of white blood cell your blood is low in:

If your blood is low in neutrophils, you have a type of leukopenia known as neutropenia. Neutrophils are the white blood cells that protect you from fungal and bacterial infections. Leukopenia is so often caused from a decrease in neutrophils that some people use the terms “leukopenia” and “neutropenia” interchangeably.

Another common type of leukopenia is lymphocytopenia, which is when you have too few lymphocytes. Lymphocytes are the white blood cells that protect you from viral infections.

# LEUKOCYTOSIS

When the number of white cells in your blood is higher than normal, it's called leukocytosis. This usually happens because you're sick, but sometimes it's just a sign that your body is stressed.

**Neutrophilia.** This is an increase in WBCs called neutrophils. They're the most common type of WBCs, accounting for 40 to 60 percent of your WBCs. Neutrophilia is the type of leukocytosis that occurs most often.

**Lymphocytosis.** About 20 to 40 percent of your WBCs are lymphocytes. An increased number of these cells is called lymphocytosis. This type of leukocytosis is very common.

**Monocytosis.** This is the name for a high number of monocytes. This cell type makes up only about 2 to 8 percent of your WBCs. Monocytosis is uncommon.

**Eosinophilia.** This means there are a high number of cells called eosinophils in your blood. These cells make up about 1 to 4 percent of your WBCs. Eosinophilia is also an uncommon type of leukocytosis.

**Basophilia.** This is a high level of WBCs called basophils. There aren't many of these cells in your blood — only 0.1 to 1 percent of your WBCs. Basophilia is rare.

# White blood cells



- **Definition:**
- **White blood cells** or **leukocytes** are cells of the immune system which defend the body against both infectious disease and foreign materials.
- **Characters of WBCs:**
  1. Whenever a germ or infection enters the body the white blood cells have a variety of ways by which they can attack. Some will produce protective antibodies that will overpower the germ. Others will surround and devour the bacteria.
  2. The white blood cells have a rather short life cycle, living from a few days to a few weeks.
  3. Several different and diverse types of leukocytes exist, but they are all produced and derived from a multipotent cell in the bone marrow known as a hematopoietic stem cell.

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## Characters of WBCs



4. Leukocytes are found throughout the body, including the blood and lymphatic system.
5. The name "White Blood Cell" derives from the fact that after centrifugation of a blood sample, the white cells are found in the Buffy coat, a thin layer of nucleated cells between the sedimented red blood cells and the blood plasma, which is typically white in color. The scientific term *leukocyte* directly reflects this description, derived from Greek *leuko* - white, and *cyte* - cell.

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## ) White cell count (WBC



- White cell count (WBC) is the total number of leukocytes in a volume of blood, expressed as thousands/ $\mu\text{l}$ .
- As with the RBC, the WBC can be done by manual methods or by automated cell counters.
- **Normal Values:**
  - Newborn 9.0-30.0 x  $10^3/\mu\text{l}$
  - 1 week 5.0-21.0 x  $10^3/\mu\text{l}$
  - 1 month 5.0-19.5 x  $10^3/\mu\text{l}$
  - 6-12 months 6.0-17.5 x  $10^3/\mu\text{l}$
  - 2 years 6.2-17.0 x  $10^3/\mu\text{l}$
  - Child/adult 4.8-10.8 x  $10^3/\mu\text{l}$

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# Leukocytosis



- Leukocytosis is a condition characterized by an elevated number of white cells in the blood, which is usually due to:
  - Bacterial infection such as appendicitis, tonsillitis, ulcers and urinary tract infection
  - Leukemia.
  - Pregnancy.
  - Hemolytic disease of new born.
  - Following exercise.
  - Emotional stress.
  - Food intake.

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# Leukopenia



- Leukopenia is a condition characterized by a decreased number of white cells in the blood, which is usually due to:
  - Viral disease such as measles and infectious hepatitis.
  - Some bacterial infections such as typhoid fever, brucellosis, and typhus fever.
  - Rheumatoid arthritis.
  - Systemic Lupus Erythematosus.
  - Certain drugs such as radio therapy and chemotherapy.

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## Principle of WBCs count test

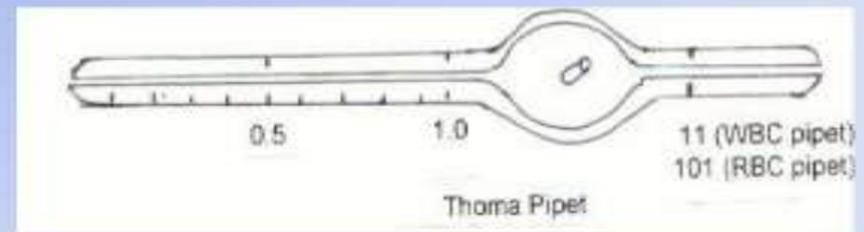


- Free-flowing capillary or well-mixed anticoagulated venous blood is added to a diluent) at a specific volume in the thoma pipette.
- The diluent lyses the erythrocytes but preserves leukocytes and platelets.
- The diluted blood is added to the hemacytometer chamber.

## Equipment



1. White blood cells count diluting fluid
2. Thoma white pipette
3. Hemacytometer and coverslip
4. Microscope
5. Lint-free wipe
6. Alcohol pads

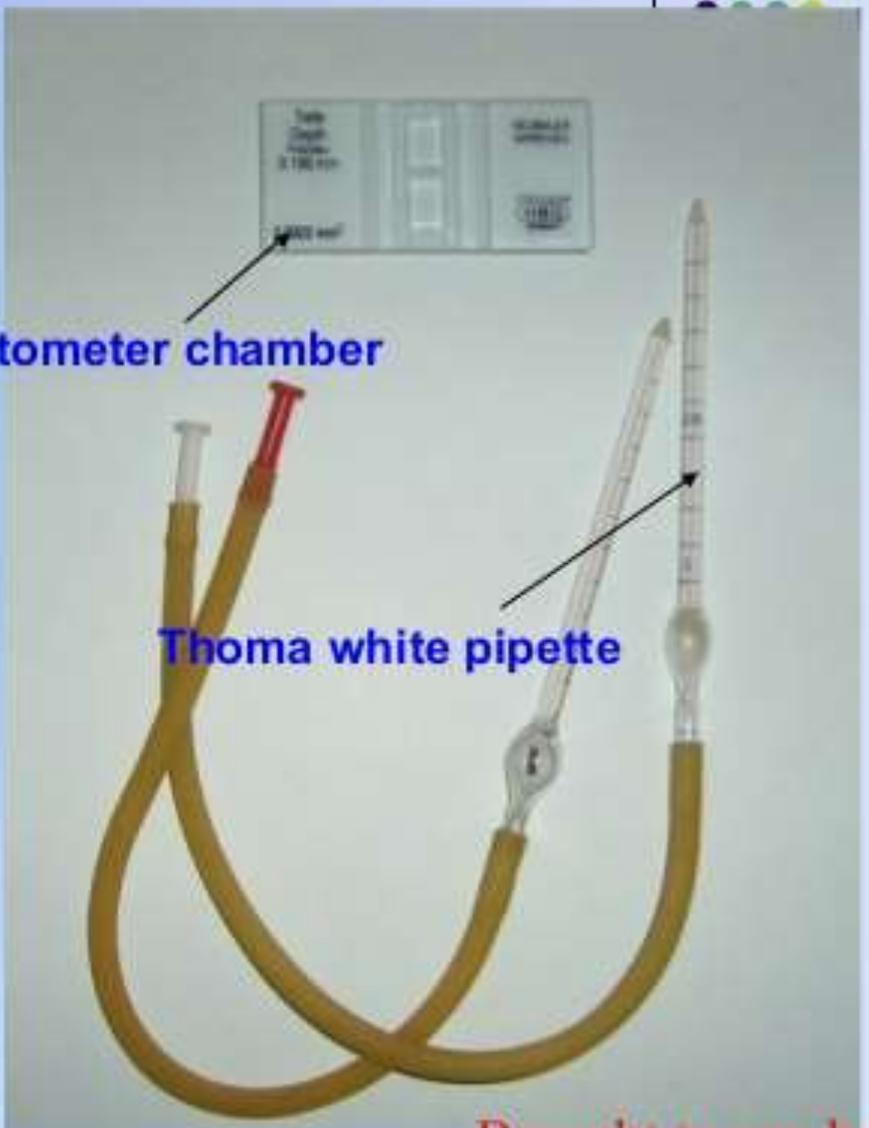


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HAEMOCYTOMETER

Rubber sucking tube



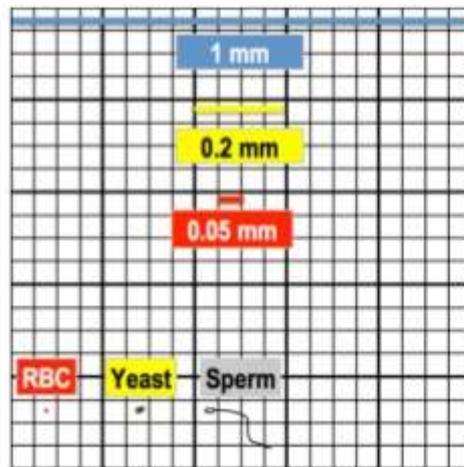
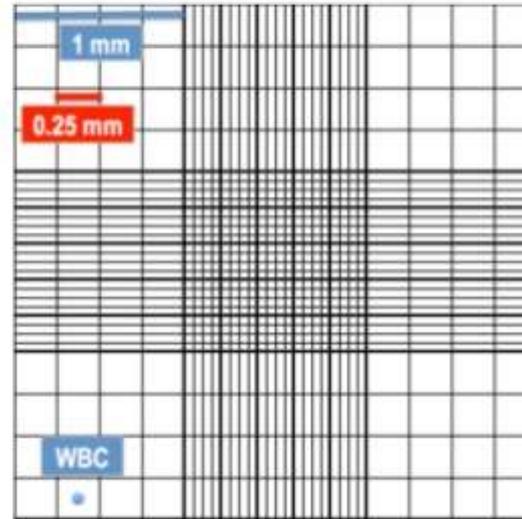
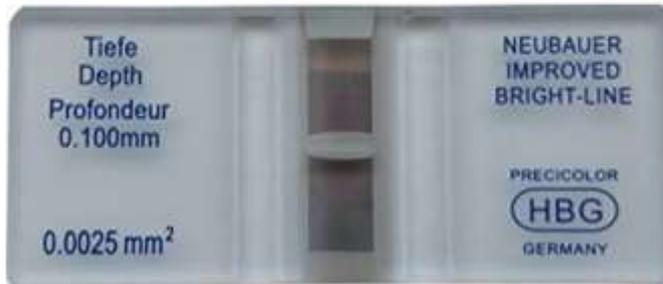
haemocytometer chamber

Thoma white pipette

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## Total Erythrocyte Counting



# Hemocytometer



- The hemacytometer counting chamber is used for cell counting.
- It is constructed so that the distance between the bottom of the coverslip and the surface of the counting area of the chamber is 0.1 mm.
- The surface of the chamber contains two square ruled areas separated by an H-shaped moat.

## Procedure



1. Draw the blood up to 0.5 mark in the thoma pipette.
2. Wipe the outside of the capillary pipette to remove excess blood that would interfere with the dilution factor.
3. Holding the pipette almost vertical place into the fluid. Draw the diluting fluid into the pipette slowly until the mixture reaches the 11 mark, while gently rotating the pipette to ensure a proper amount of mixing.
4. Place the pipette in a horizontal position and firmly hold the index finger of either hand over the opening in the tip of the pipette, detach the aspirator from the other end of the pipette now the dilution of the blood is completed

## Procedure



5. Mix the sample for at least 3 minutes to facilitate hemolysis of RBCs.
6. Clean the hemacytometer and its coverslip with an alcohol pad and then dry with a wipe.
7. Before filling the chamber, discard the first four to five drops of the mixture on a piece of gauze to expel the diluent from the stem.



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## Procedure



8. Carefully charge hemacytometer with diluted blood by gently squeezing sides of reservoir to expel contents until chamber is properly filled.



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## Procedure for counting WBC's



1. Under 10 x magnifications, scan to ensure even distribution. Leukocytes are counted in all nine large squares of counting chamber.
2. Count cells starting in the upper left large corner square. Move to the upper right corner square, bottom right corner square, bottom left corner square and end in the middle square.
3. Count all cells that touch any of the upper and left lines, do not count any cell that touches a lower or right line.

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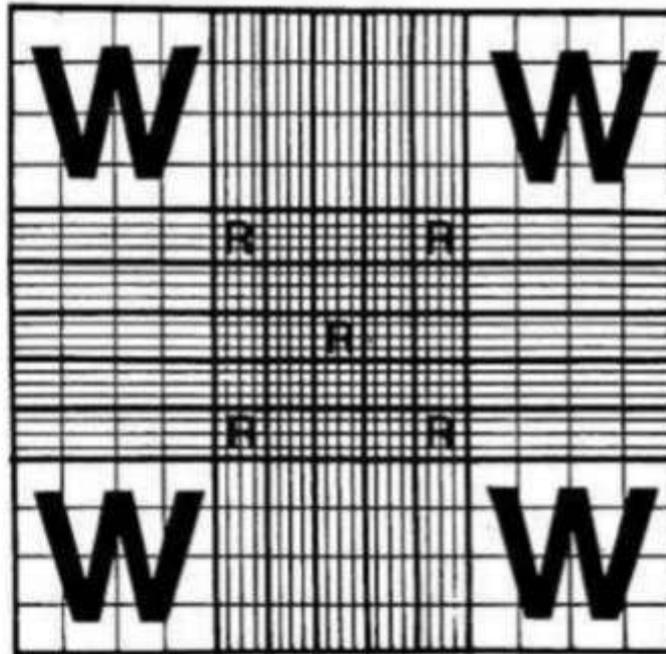
Small square = 1/400 sq. mm.    1/25 sq. mm.

← 1 millimeter →

Counting grid (central area)

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## Total Erythrocyte Counting





**THANKS**