

EXPERIMENT NO.: 8

DATE:

AIM: TO ESTIMATE DIFFERENTIAL WBC (LEUKOCYTE) COUNT OF OWN BLOOD SAMPLE

REQUIRMENT:

Microscope, Thomas Coverslip, Microscope, Cotton swab, Pricking Needle/Lancet, 70 % Methylated Alcohol (Spirit), Giemza or Leishman stain.

THEORY:

White blood cells (leukocytes) have two types:

a. Granulocytes (BEN):

- 0.5-1.0% Basophils
- 2-4% Eosinophils
- 60-70% Neutrophils

b. Agranulocytes (LM):

- 20-25% Lymphocytes
- 3-8% Monocytes

Granular leukocyte:

- It develops from myeloblast.
- It contains protein which is known as major histocompatibility (MHC) antigen.
- It contains the clear granules in cytoplasm that can be seen under light microscope.
- It is further divided into three types:

a) Eosinophils:

- It is 10-14 μm in diameter.
- Its granules produce red or orange stain with acidic dyes.
- The nucleus of eosinophils has two lobes connected by thin or thick fiber.
- The granules are large and uniform in size that are present in group in cytoplasm but do not cover the nucleus.

b) Basophils:

- It is 8-10 μm in diameter.
- Its granules give Blue-purple stain with basic dyes.
- Its nucleus is in irregular shape often in form of letter S.
- The cytoplasmic granules are round and variable in size.

c) Neutrophils:

- It is 10-12 μm in diameter.
- Its granules known as neutral because it produces pale lilac stain with mixture of acidic and basic dye.
- Their nuclei contain two to five lobes connected by very thin fibers of chromatin.
- Older neutrophils known as polymorphonuclear leukocytes (PMNs), polymorphs or polys because it has many different shaped nuclei.
- Younger neutrophils are known as bands because their nucleus is rod shaped.

2) Agranular leukocyte:

- It has the granules but do not seen under the light microscope because of their small size so it is known as agranular.
- It is further classified into two types:

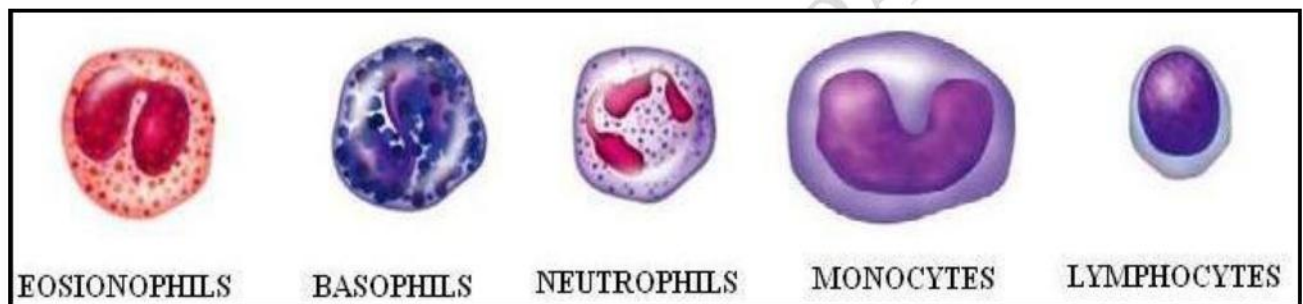
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a) Lymphocytes:

- Small lymphocytes are 6-9 μm in diameter and large lymphocytes are 10-14 μm in diameter.
- It is develop from lymphoblast.
- Their nucleus is in round shaped and produce dark stain.
- The cytoplasm produce border around the nucleus and it produce sky blue stain.

b) Monocytes:

- It is 12-20 μm in diameter.
- It is develop from monoblast.
- Their nucleus is in kidney shaped.
- Its cytoplasm has foamy appearance and produce blue grey stain.
- Monocytes migrate from blood to tissue where they enlarge their size and differentiated in to macrophages.
- Some are known as fixed macrophages because they are fixed on particular tissue such as alveolar macrophages, spleen macrophages etc.
- Other are known as free or wandering macrophages which travel tissue to tissue at inflammation or infection site for to repair it.



PROCEDURE:

- Place drop of blood on dried slide.
- Spreading it with another slide (blood film method).
- Add Giemza or Leishman stain or Field Stain or Wright Stain (Any One) on to the blood film.
- Allow to stain for 2-3 min.
- Wash gently in a stream of buffered water until it has acquired a pinkish tinge (up to 2 min).
- Observe the different types of WBC in Microscop first in 40X or 45X then by oil immersion lens – 100X) and record the number of all WBC types that observed.

N	L	M	L	E					

Symbols to be used:

- 'M' for Monocyte
- 'L' for Lymphocyte
- 'E' for Eosinophil
- 'B' for Basophil
- 'N' for Neutrophil

Fill these Symbols accordingly in the 100 squares on the left

OBSERVATION TABLE

Result:

Types of Cell	Total Number of Cell	Amount of Cell in %
Basophil		
Eosinophil		
Neutrophil		
Lymphocyte		
Monocyte		

CONCLUSION: My different types of WBCs are in **normal** / **abnormal** range.

DISCUSSION:

Type of white blood cell	Function
neutrophil	helps stop microorganisms in infections by eating them and destroying them with enzymes
lymphocyte	–uses antibodies to stop bacteria or viruses from entering the body (B-cell lymphocyte) –kills off the body's cells if they've been compromised by a virus or cancer cells (T-cell lymphocyte)
monocyte	becomes a macrophage in the body's tissues, eating microorganisms and getting rid of dead cells while increasing immune system strength
eosinophil	helps control inflammation, especially active during parasite infections and allergic reactions, stops substances or other foreign materials from harming the body
basophil	produces enzymes during asthma attacks and allergic reactions

INTERPRETATION:

An **increased percentage of neutrophils** in your blood can mean that you have:

- Neutrophilia, a white blood cell disorder that can be caused by an infection, steroids, smoking, or rigorous exercise
- an acute infection, especially a bacterial infection
- acute stress
- pregnancy
- inflammation, such as inflammatory bowel disease or rheumatoid arthritis
- tissue injury due to trauma
- chronic leukemia

A **decreased percentage of neutrophils** in your blood can indicate:

- neutropenia, a white blood cell disorder that can be caused by a lack of neutrophil production in the bone marrow
- aplastic anemia, a decrease in the number of blood cells produced by your bone marrow
- a severe or widespread bacterial or viral infection
- recent chemotherapy or radiation therapy treatments

An **increased percentage of lymphocytes** in your blood may be due to:

- lymphoma, a white blood cell cancer that starts in your lymph nodes
- a chronic bacterial infection
- hepatitis
- multiple myeloma, a cancer of the cells in your bone marrow
- a viral infection, such as mononucleosis, mumps, or measles
- lymphocytic leukemia

A **decreased percentage of lymphocytes** in your blood can be a result of:

- bone marrow damage due to chemotherapy or radiation treatments
- HIV, tuberculosis, or hepatitis infection
- leukemia
- a severe infection, such as sepsis
- an autoimmune disorder, such as lupus or rheumatoid arthritis

A **heightened percentage of monocytes** in your blood can be caused by:

- chronic inflammatory disease, such as inflammatory bowel disease
- a parasitic or viral infection
- a bacterial infection in your heart

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- a collagen vascular disease, such as lupus, vasculitis, or rheumatoid arthritis
- certain types of leukemia

An **increased percentage of eosinophils** in your blood can indicate:

- eosinophilia, which can be caused by allergic disorders, parasites, tumors, or gastrointestinal (GI) disorders
- an allergic reaction
- skin inflammation, such as eczema or dermatitis
- a parasitic infection
- an inflammatory disorder, such as inflammatory bowel disease or celiac disease
- certain cancers

An **increased percentage of basophils** in your blood might be caused by:

- a serious food allergy
- inflammation
- leukemia

SIGNATURE OF TEACHER