Differential leucocytes count

Aim: To enumerate the different types of leucocytes namely neutrophil, eosinophil, basophil, monocyte, and lymphocyte.

Method: Blood film (smear) with Wright's or Giemsa stain

Significance

- They are responsible for the defense of the organism
- They are 4000-10000mm3
- Leucocytes divide in to granulocyte and agranulocyte

Neutrophils

Very active in phagocyting bacteria, found in highly in pus, Died after phagocyting pathogens and high during inflammation.

Eosinophils

Attack parasites and phagocyte antigenantibody complexes.

Basophils

Secrete anticoagulant and vasodilatory substances (histamines, serotonin)

Lymphocytes

The main constituents of immune system defense against viruses, bacteria, fungi and protista.

Monocytes

Precursors of macrophages. Have intense secretory activities (lysosomes, interferons)

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Blood Cells					
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Monocyte	Lymphocyte	Neutrophil	Eosinophil	Basophil	
			Ale 6		
Macrophage		Erythrocyte	Platelets		



Requirements

Clean glass slide, sterile needle, microscope, stains solution, oil immersion

Protocol

- 1. Making the smear
- 2. Place a small drop of blood near an end of the slide
- Bring another slide in contact with the drop and allow distributing at an angle of 30-40 degrees
- 4. Push to the left in a smooth and quick motion
- 5. Dry the slide with air
- 6. Fixing Dip the smear in a vessel containing 95% containing ethyl or methyl alcohol for 3-5 minutes
- 7. Stain the smear with Giemsa or Wright's stain for 16-60 or 1-3 minutes respectively
- 8. Rinse the slide with distilled water at room temperature
- 9. Drain off the water and leave the slide to dry
- 10.Place a drop of oil immersion on the smear and then mount the cover slip

Observation

- 1. examine under dry objective or oil immersion
- 2. count about 100-200 cells and take average

Interpretation

Neutrophils

- Very tiny light staining granules
- Multi-lobed nucleus connected by thin strands of nuclear material
- Are most numerous, normally they account 50-70%



• If the count exceeds this amount, the cause is usually due to an acute infection

(appendicitis, small pox, rheumatic fever)

- If the count is considerably less, it may be due to a viral infection (influenza, hepatitis, rubella).
- The typical response to infection or serious injury is an increased production of

Eosinophils

- Has large granules (acidophilic), pink red
- The nucleus often has two lobes connected by a band of nuclear material, looks like telephone receiver which contains digestive enzymes against parasites.

Elevations in eosinophil counts are associated with:

- Allergic reactions
- Parasite infections
- Chronic skin infections
- Some cancers

Decreases in eosinophil counts are associated with:

- Stress
- Steroid exposure
- Anything that may suppress WBC production generally

Basophils (Baso's)

These cells can digest bacteria and other foreign bodies (phagocytosis) and also have some role in allergic reactions.

Elevations in basophil counts are associated with:

- Some cancers
- Some allergic reactions





Figure 2:. Eosinophil has granules



- Some infections
- Radiation exposure

Diminished basophil counts are associated with:

- Stress reactions
- Some allergic reactions
- Hyperthyroidism
- Prolonged steroid exposure

Lymphocytes

- An agranulocyte cell with very clear cytoplasm Very large nucleus which fills almost the cell with leaving thin rim of cytoplasm
- Smaller than the three granulocytes
- Play an important role in our immune response
- The T-lymphocytes act against virus infected cells and tumor cells
- .Figure 3: Normal lymphocytes The B-lymphocytes produce antibodies and numerous leukocyte, accounting for 25-35% of the counted in differential WBC count.
- When number of this cells exceeds the normal value, one would suspect parasites
- Patients with AIDS keep a careful watch on their T-cell level

increased numbers of lymphocytes are seen in:

- Most viral infections
- Some bacterial infections
- Some cancers
- Graves' disease

Decreased numbers of lymphocytes are seen in:

- Steroid exposure
- Some cancers







- Immunodeficiency
- Renal failure
- Lupus

Monocytes

- The largest of leucocytes and are agranular
- "U" or kidney bean shaped nucleus
- Abundant cytoplasm and light blue
- Account for 3-9% of the total leukocyte

Increased monocyte counts are associated with:

- Recovery from an acute infection
- Viral illness
- Parasitic infections
- Collagen disease
- Some cancers
- People with malaria, endocarditis, typhoid fever, and rocky mountain spotted fever, monocyte increases in number

Decreased monocyte counts are associated with:

- HIV infection
- Rheumatoid arthritis
- Steroid exposure
- Some cancers

Normal value

Neutrophil = 50-70%	Eosinophil = 2-4%	Basophil = $0-1\%$
Lymphocytes = 20-40%	Monocytes $= 3-8\%$	

References:

1. Jelalu Kemal .(2014).Laboratory Manual and Review on Clinical Pathology.. OMICS Group eBooks,USA.

