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### Diploma in Pharmacy 1<sup>st</sup> Year Human Anatomy & Physiology Experiment

#### To determine the differential count of blood.

### Aim:

To determine the differential count of blood.

### **Reference** :

Dr. Gupta G.D , Dr. Sharma Shailesh , Dr. Sharma Rahul Kumar , "Practical Manual of Human Anatomy and Physiology" Published by Nirali Prakashan , Pg.No 77 - 80

## Material and Apparatus Required

Cotton, spirit, needle, glass slide, distilled water, Leishman's stain, light microscope with oil immersion objective (90x).

# Theory :

- A differential blood count gives an estimate of relative percentage of each type of white blood cell. It also aids in diagnosing abnormal white blood cell populations, like blasts, immature granulocytes, and circulating lymphoma cells in the peripheral blood.
- 4,500 to 11,000 WBCs per microliter (4.5 to 11.0 x 109/L) is the normal number of WBCs in the blood. This range may vary slightly among different labs. Normal range of neutrophil is 2500-8000 per mm (55-70%).

## Procedure

### 1) Preparing the Blood Smear:

- i. The fingertip should be sterilised with a cotton swap dipped in 70% alcohol and should be dried.
- ii. A bold prick should be made on the fingertip for the free flow of the blood.
- iii. The drops of blood should be collected on the end side of glass slide.

- iv. The blood drop should be spread with another glass slide by placing it a an angle of 45 degree and moving it sidewise.
- v. The spreader should be held firmly and moved on the previous slide to
- vi. the other end in a straight line with same force and pressure.
- vii. The glass slide should be allowed to dry after formation of the smear.

#### 2) Staining the Slide:

- The smeared glass slide should be kept on a flat surface with the smeared surface facing upwards.
- The drops of Leishman's stain should be poured on the glass.
- Then the stain drop should be slid to cover the smear film.
- It should be kept undisturbed for 2-5mm.
- The drops of distil water should be poured on the slide and left for 10 minute.
- The dye and water should be removed. The stain should also be removed by keeping the slide under running water.
- The slide should he kept aside for some time to dry.

### 3) Observation of the Glass Slide and Counting of Cells:

- The prepared glass slide should be kept under low power of compound microscope and a good quality slide should be chosen.
- Then different types of WBC should be identified under medium power.
- A table should be drawn with 10 boxes both on horizontal and vertical axis on an observation notebook.
- The slide should be fixed on the platform and an area towards the corner should be chosen
- The different types of WBC found should be noted on the table in on abbreviated
- Should be moving downwards and in chain like manner till 100 cells are observed.
- The report should be prepared after counting 100 cells



### **Representative Example**

L	M	N	L	N	L	N	L	L	N
N	N	N	N	L	N	N	N	M	N
L	N	L	L	N	N	M	N	L	L
M	N	N	L	L	M	N	M	L	N
L	M	N	N	N	L	L	N	Ν	Μ
Μ	N	N	N	L	L	N	N	L	N
L	N	N	L	L	N	N	L	L	N
Μ	Ν	L	L	N	L	M	N	N	B
Μ	L	L	Ν	N	N	N	N	L	N
Ν	L	L	L	N	N	N	N	N	E

Where

N-Neutrophils, B-Basophils L-Lymphocytes E-Eosinophils M-Monocytes

#### **Observation Table**

The total count of every class should be made to determine its percentage

(%)

**Observation Table** 

The total count of every class should be made to determine its percentage (%).

Serial Number	Types of Cells	Count	Total
1)	Neutrophils		65
2)	Eosinophils	III	03
3)	Basophils		0.0
4)	Monocytes	IIII	04
5)	Lymphocytes		28
	+	Total	100

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## **Result:** Differential count of given sample of blood is as follows:

Cells	Percentage(%)
Neutrophils	
Eosinophils	
Basophils	
Monocytes	
Lymphocyte	

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