Precautions

1. There should be no air bubble in the column of the blood.
2. The cells should be evenly distributed.

Calculation

No of cells in four large WBC square	= x
No of cells in one WBC square	= x/4
Side of 1 WBC square	= 1 mm
Area of 1 WBC square	= 1 mm^2
Depth of chamber	= 1/10 mm
Volume of fluid present over 1 WBC square	= 1/10 mm^3
No of cells in 1/10 cu mm of diluted blood	= x/4
No of cells in 1 cu mm of diluted blood	= x/4 × 10
Dilution factor	= 1:20
No of cells in 1 cu mm of undiluted blood	= x/4 × 10 × 20
	= 50x

DISCUSSION

(Normal range of WBC is 4000–11000 cu mm in one) Total leucocytes count shows variation under certain physiological and pathological conditions.

Leucocytosis (increase in WBC Count)

Physiological Conditions

i. Age
   a. Newborn will always have an increased count.
   b. Childhood, pregnancy and delivery shows increased count.
   c. There is no significant change in old age compared to adult values.
ii. Females during pregnancy and parturition.
iii. Stress like severe exercise, severe pain, and excitation.
iv. Diurnal variation: WBC count may vary from hour to hour. Highest count in evening and lowest count in morning).
v. Digestive leucocytosis (after digestion).
vi. Injection of adrenaline.

Pathological Conditions

a. *Acute pyogenic infections:* For example, pneumonia, appendicitis and tonsillitis.
b. *Leukaemia (abnormal increase with immature cells):* Count may go up to 1,00,000 to 3, 00,000 per cu mm.
c. Acute haemorrhage.
d. Tissue damage resulting from burns, operations, myocardial infarction.
e. Malignant neoplasms.
f. Metabolic disorder—For example, gout, diabetic acidosis

Leucopenia (decrease in WBC count)

Physiological Conditions

Rare but sometimes due to:
1. Exposure to cold
2. Aspirin.

Pathological Conditions

a. Infection—typhoid.
b. Starvation—malnutrition.
c. Viral infections—measles, chickenpox, influenza, rubella
d. Drugs—antimetabolites, antimicrobials (sulphonamides, chloramphenicol).
e. Haematological disorders—aplastic anaemia, pernicious anaemia, irradiation).

QUESTIONS

1. Is this a routine test? What is the usefulness of this test?
2. What other test has to be done with total count to make it more meaningful?
3. Why should you destroy the RBCs while doing WBC count?
4. What is the normal range of total WBC count? List 3 important physiological variations?

nuclear lobes joined by chromatin strands should be a granulocyte. A big cell with a kidney-shaped nucleus is a monocyte. The nucleus in a lymphocyte completely fills the cells.

Cytoplasm

a. The amount of cytoplasm in relation to the size of the nucleus is noted.
b. The presence of visible granules and their nature is observed.

A small cell with a thin crescent of cytoplasm is a small lymphocyte. A bigger cell with a rim of cytoplasm all around is a large lymphocyte. A big cell with large amount of clear cytoplasm in relation to the nucleus is a monocyte.

If a granulocyte has fine neutral granules of light violet colour, the cell is a neutrophil. If the granules are coarse and orange or red coloured, the cell is an eosinophil. In a basophil, the granules are large, coarse and deep blue in colour. It is the smallest of all granulocytes.

Results

Neutrophils	%
Eosinophils	%
Basophils	%
Lymphocytes	%
Monocytes	%

DISCUSSION

The percentage of the different types of white blood cells is called the differential count. It is done to find out if there is an increase or decrease in a particular type of leukocyte. But it shows only a relative increase or decrease of the cell type with a corresponding change in the other cell types. Absolute values are more significant relative value.

Normal Count

Neutrophils	50–70%
Eosinophils	1–4%
Basophils	0–1%
Lymphocytes	20–40%
Monocytes	2–8%

Causes

Neutrophilia

It is an increase in the number of neutrophils:

1. Acute pyogenic infection such as tonsillitis, appendicitis, pneumonia
2. Tissue necrosis as in myocardial infarction
3. Following haemorrhage
4. Trauma, postoperative burns
5. Haemolysis
6. Malignant neoplasm
7. Metabolic disorders like gout, diabetic acidosis, uremia
8. Drugs such as glucocorticoids, adrenaline, digitalis, phenacetin
9. Poisoning with lead, mercury, insect venom.
10. Physiological-exercise, stress, after meals, pregnancy and parturition.

Neutropenia

Neutropenia is decrease in the number of neutrophils:

1. Typhoid and paratyphoid fever, kala azar
2. Viral infection
3. Depression of bone marrow due to irradiation,

Cell type	*Size*	*Nucleus*	*Cytoplasm*
Granulocyte			
Neutrophil	10–14 μm	2–6 lobes connected by chromatin	Fine light violet coloured granules
Eosinophil	10–14 μm	Bicobed connected by a chromatin strand (spectacle-shaped)	Coarse orange to brick red granules.
Basophil	10–14 μm	Irregular may be bilobed or S-shaped. Not seen clearly as it is obscured by the granules.	Coarse deep blue granules completely filling the cell.
Agranulocytes			
Lymphocyte			
Small	8–10 μm	Large-round nucleus completely, filling the cell, stains deep blue.	Thin crescent of pale blue cytoplasm. No granules seen
Large	10–15 μm	Large round or indented nucleus, stains deep blue	Thin rim of pale blue cytoplasm all around. No granules seen
Monocyte	16–24 μm	Large, placed centrally, kidney-shaped nucleus	Large amount of pale grey blue cytoplasm. No granules seen.

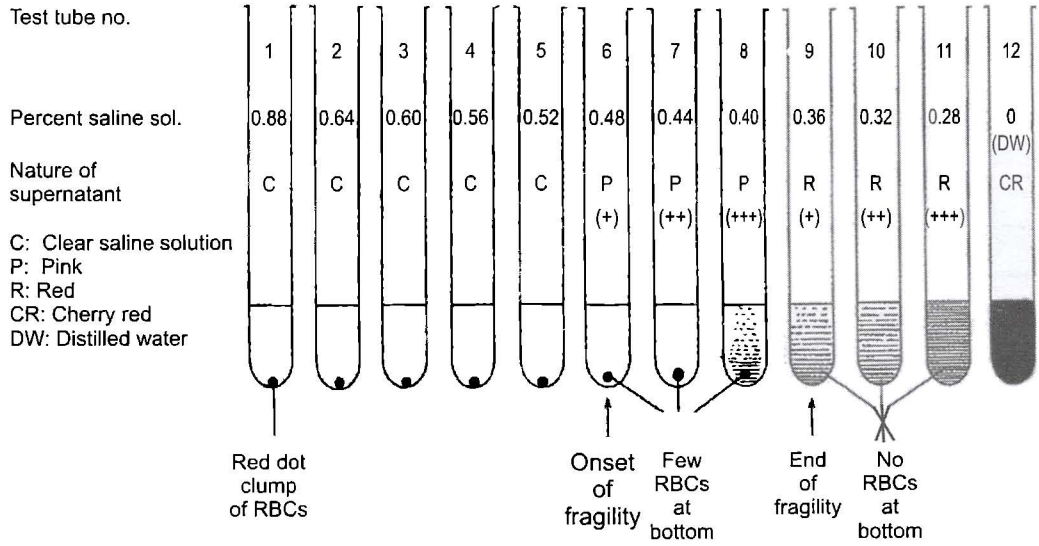

Fig. 13.1: Unit 1: Haematology

2. RBC's osmotic fragility increases when it becomes spherical, e.g. in spherocytosis (independent of the cause). It begins at 0.7% NaCl and ends at 0.45% NaCl solution.
3. RBC's osmotic fragility is decreased when it becomes slender, e.g. in iron deficiency anaemia and in thalassaemia. It begins at 0.36% NaCl and ends at 0.24% NaCl solution.

Result

Haemolysis begins at .....% saline and ends at .....% saline.

QUESTIONS

1. Would you say the fragility is increased or decreased in the following situations?
   a. Haemolysis begins at 0.55 g% and is complete at 0.45 g%
   b. Haemolysis beings at 0.3 g% and is completed at 0.25 g%.
2. Name one condition where fragility is increased and say why?
3. Name one condition where fragility is decreased.
4. Name one other aetiology where fragility is increased in the RBC.
5. What is the difference between isotonic and iso-osmotic solutions?
6. Define the terms haemolysis and crenation: When do they occur?
7. Name some haemolytic agents.
8. What is mechanical fragility and how is it tested?
9. Do all the normal RBCs present in the given sample of blood have the same osmotic fragility to hypotonic solutions?
10. What happens to the osmotic fragility of RBCs in an individual after splenectomy?
11. Define the terms osmosis and osmotic pressure. How much osmotic pressure is exerted by the blood and what is its importance in the body?
12. Explain the mechanism of haemolysis of RBCs of the body?

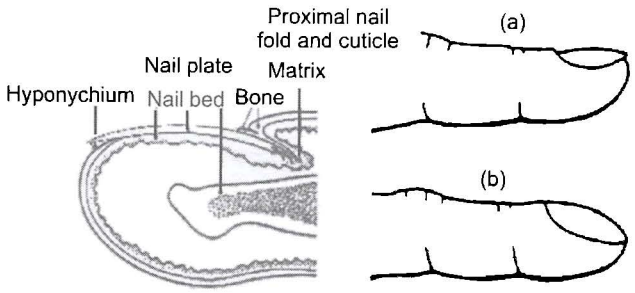

Fig. 1.1: Anatomy of nail: (a) Normal, (b) Clubbing

- Rhythm—the regularity with which one pulse follows the other.
- Volume—the amplitude of expansion of vessel wall.
- Character—whether collapsing or not (rise, maintenance and fall); the contour of a pulse.
- Condition of vessels wall—thickened or not.
- Other peripheral pulsations, i.e. brachial, carotid, facial, femoral, popliteal, dorsalis pedis and posterior tibial.