

# **The red blood cell (RBC) count**

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# The red blood cell (RBC) count

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The number of red blood cells per unit volume of whole blood.

- Normal range of RBCs Adults : 4.8-7.2 million (male) and 4.9-5.5 million (female).

Pregnancy: slightly lower than normal adult values. Children : 3.8-5.5 million.

The number of RBCs varies with age, sex, and altitude.

- Each RBC has a mean diameter of about 7.2  $\mu\text{m}$  and thickness of 2.5  $\mu\text{m}$  at the thickest point and 1  $\mu\text{m}$  or less at the center .
- The main constituent of the RBCs is hemoglobin which enables them to transport oxygen around the circulation .

# Medical condition

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Medical condition in which a decrease in RBC count can be found is **anemia**.  
The condition of increased number of RBC is called **polycythemia**.

A lower than normal RBC can result from a number of causes including:

Massive RBC loss, such as acute hemorrhage

Abnormal destruction of red blood cells

Lack of substances needed for RBC production

Chemotherapy or radiation side effects from treatment of bone marrow malignancies such as leukemia can result in bone marrow suppression.

# Manual RBC count Materials

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1. Anticoagulated whole blood (using **EDTA** or **heparin** as an anticoagulant) or capillary blood can be used.

2. **Hayem's** solution (diluting fluid) composed of:

❖ Sodium chloride (NaCl) – 0.5 g to maintain isotonicity

❖ Sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>) – 2.5 g to prevent Rouleaux formation

❖ Mercuric chloride (HgCl<sub>2</sub>) – 0.25 g to fix the cells and act as a preservative.

❖ Distilled water – 100 mL

**RBC pipette:** which is composed of a stem & a mixing chamber with a red bead, its function is to mix blood with the substance and for differentiation from the WBC pipette.

4. **hemocytometer** chamber

5. Microscope, Lancet, Alcohol 70% and cotton

# Manual RBC Count Materials and Instruments



**RBC Diluting Fluid**



**Microscope**



**RBC Pipette**

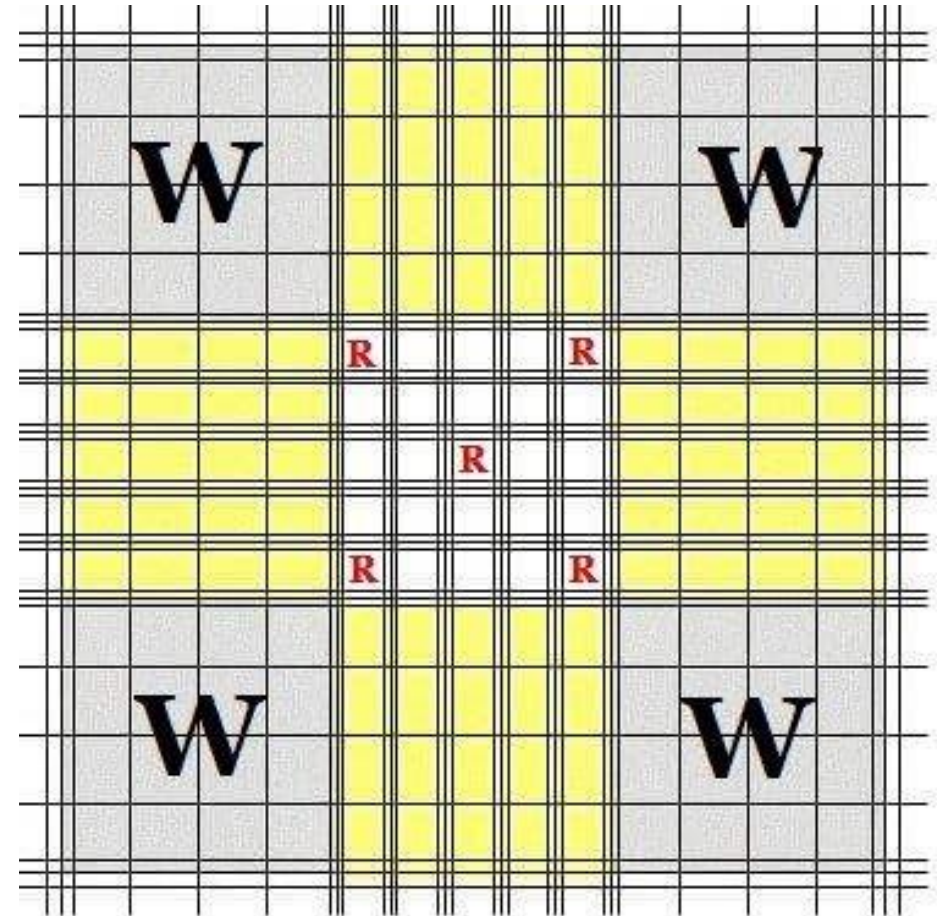
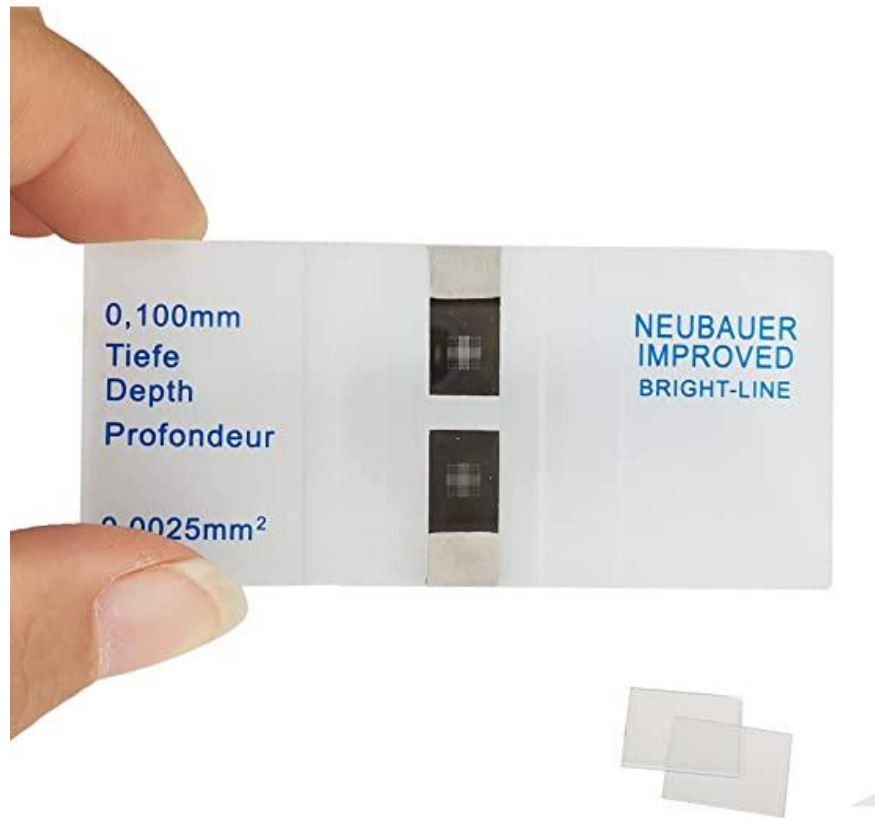


**Neubauer Chamber**



**Alcohol Pad**

# hemocytometer chamber



# Procedure

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1. With a sterile disposable lancet do small prick on the finger tip, hold the red cell pipette slightly tilted from the vertical position, apply its tip to the drop and aspirate blood to the **mark 0.5**.
2. Wipe off any blood adhering to its outer side. If the blood gets beyond 0.5 marks tap the tip gently till the blood is exactly at the mark. Never allow the blood to clot inside the pipette. If the blood clots in the pipette blow the sample out, clean the pipette and begin all over again.
3. Aspirate diluting **Hayem's** solution to the 101 mark, thus making 1:200 dilution of blood.
4. Hold the pipette horizontally and roll it with both hands between finger and thumb.

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1. Blow out a **quarter** of the contents to remove the pure diluting fluid in the stem.
  2. Prepare the counting chamber and cover it with a cover slip. Hold the pipette and Touch its tip gently on the surface of the counting platform and squeeze to apply a drop of mixture.
  3. Place the **hemocytometer** chamber on the stage of the microscope and allow 2 minutes for the cells to settle.
  4. Scan the counting area with 10x objective lens. Use the 40X objective, include all cells lying on the lower and left lines of any square; omit the cells on the upper & right hand lines. Count the cells in 5 medium squares of 16 small square i.e. 80 small squares, one at each corner and one in the center.



# Calculation

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Count the number (N) of cells in 80 small squares located in 5 middle-sizes squares (four located at the four corners and one in the middle). The size of 80 small squares in which “N” numbers of cells are found is:

$$1/20 \times 1/20 \times 1/10 \times 80 = 1/50 \text{ mm}^3 .$$

1/20 mm is the sideline of the square, 1/10 mm is the depth of the counting chamber between cover slip and the ruling, 80 is the number of small squares used to count.

Therefore the total numbers of cells in 1 mm<sup>3</sup> are = **N x 50** (diluted sample)

The actual total number of cells before dilution should be:

$$\mathbf{N \times 50 \times 200 = N \times 10000}$$

# Counting Chamber

