Complete blood count (CBC) – Practical-2:

It includes:

1-Red blood cell count / μ l of blood 2- White blood cell count μ l of blood. / μ l of blood. 3-packed cell volume or microhaematocrit value (PCV% or HCT %) 4- Haemoglobin concentration in gram/ deciliter (g/dl) of blood. 5-Erythrocyte indices: MCV (fl), MCH (pg) & MCHC (g/dl). 6- Differential leukocyte count. 7-Plateletes count. 8- Others.

Anemia is defined as a decrease in the hematocrit, erythrocyte count, and hemoglobin concentration. **Relative anemia** is not associated with a true reduction in the total red blood cell (RBC) mass, but rather is caused by a dilutional effect that can be observed after aggressive fluid therapy or other cause of increased plasma volume. **Absolute anemia** is considered an actual reduction in the total RBC mass.

Polycythemia is defined as an increase in the hematocrit, erythrocyte count, and hemoglobin concentration. In relative polycythemia, total erythrocyte mass is normal but appears to increase. In absolute polycythemia, increased erythrocyte production results in a true expansion of the total erythrocyte mass.

1- RBC count:

<u>Indications:</u> For the diagnosis of anaemia and polycythemia. <u>Materials:</u>

*Fresh well- mixed blood sample.

*Diluting fluid (Hayem's solution).

* RBC diluting pipette.

*Haemocytometer.

*Coverslip

*Microscope.

Method:

* Pipette well-mixed blood to the mark 0.5 as shown in the diagram.

* Wipe excess blood from outside of the pipette.

* Draw diluting fluid to the mark 101.

* Close tip of the pipette with your thump and rubber tube end with the index, mix thoroughly in figure eight for two minutes.

* Dilution will be 1:200.

* Clean the counting chamber, dry it and apply clean cover slide over the central area, apply gentle pressure on it.

* Blow out from the pipette one third of its contents to remove diluting fluid from the pipette stem.

* Holding the pipette in an angle of ^o45 to the vertical, touch tip of the pipette on the area between the cover & central area of the haemocytometer.

* A drop of diluted blood will escape and cover the graduated area. Avoid air bubble formation or escape of fluid to surrounding grooves.

*Place the haemocytometer on the microscope stage leave it for i-2 minutes to allow cell setting.

*Under the high power (X 40) count number of rbcs in 5 squares (A, B, C, D, E) each of which is surrounded by triple lines and contain 16 smaller squares (reduce intensity of illumination).

<u>Calculations:</u> Dilution factor = 1/200, depth 1/10, volume x5.

No. of RBCS /µl of blood = No of cells /5 small squares x 200 x10 x 5

(10000 - fixed factor).

=No of cells /5 small squares x10000.

Table (1) :Normal RBC count in different animal species

| Animal sp. | RBCs ×10 ⁶ /µl |
|------------|---------------------------|
| cow | 5-10 |
| Horse | 6.5-12.5 |
| Dog | 5.5-8.5 |
| Sheep | 9-15 |
| Goat | 9-15 |



Fig. 3-1. Blood dilution pipettes. A, Capable of dilutions of 1:10 and 1:20, commonly used for leukocyte dilutions; B, capable of dilutions of 1:100 and 1:200, commonly used for erythrocyte dilutions.



Figure 6-2 Neubauer hemacytometer. The large *Ws* indicate the squares that are counted for a total white blood cell (WBC count with the 1:20 dilution WBC Unopette system. The small *R* indicate the squares that are counted for a red blood cell (RBC count with the RBC Unopette system.