Hemoglobin (Hb) Estimation

Hemoglobin abbreviated Hb, is a substance which is the main constituent of the Red Blood Cells, it is the most important pigment of the blood importing red color to it.

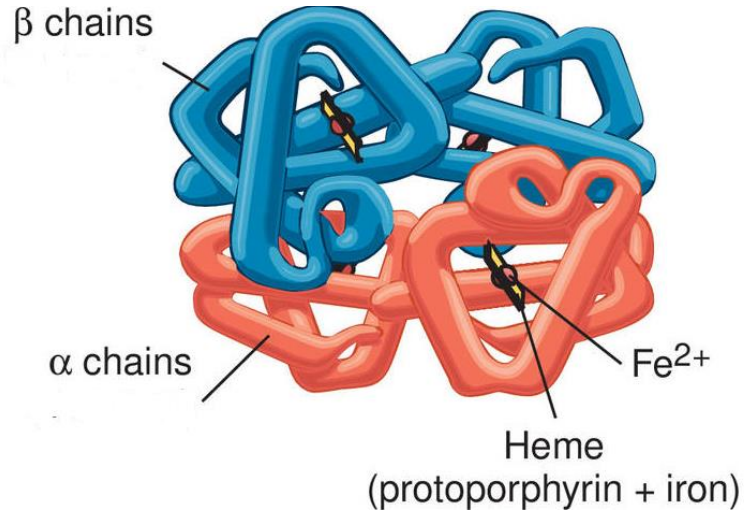
the main function of Hb is oxygen carrying from lungs to the body tissue and the CO2 transport in the blood.

Hb is a protein in nature composed of two portions:

1. Heme: A heme group consists of an iron and a porphyrin
2. Globin: which consists of two pairs of chains α and β. Globin:

There are three types of Hb:

1. Hb (HbA)**:** present in Adults.
2. Hb Portland: present in Embryonic life.
3. Hb (HbF): present in Fetal life.



Hb test :

- **Sahli's/acid hematin Method :**

Test principle

Hb in the red blood cells convert to acid haematedin by the action of HCL the brown color developed is matched against standard brown glass in the comparator by direct vision.

This method need:

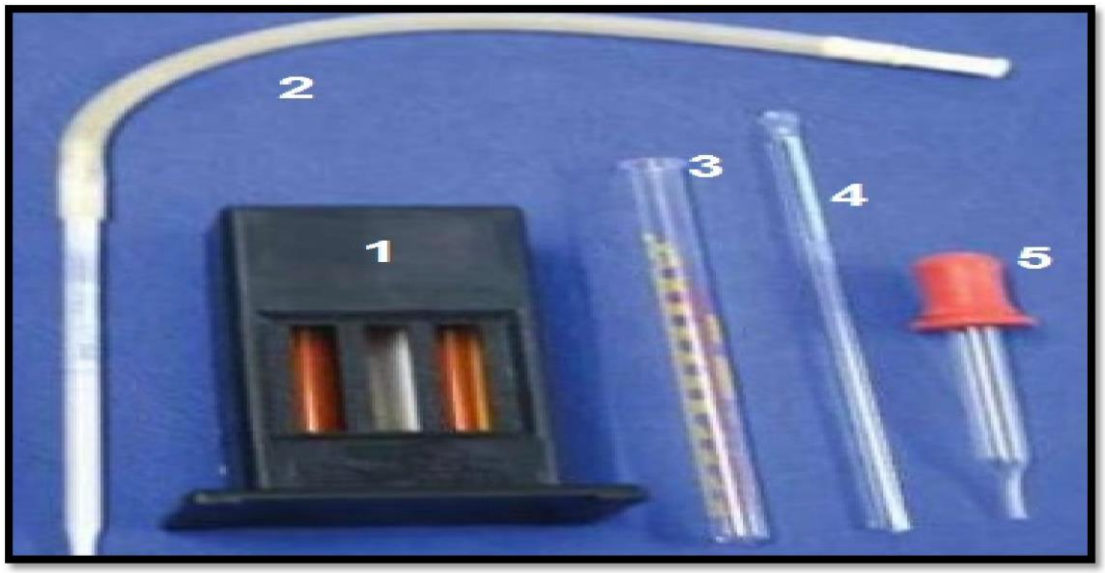
A. Blood sample (capillary or venipuncture blood collection in EDTA tube).

B. Hemocytometer , picture (1) which consists of :

1. Comparator box which has brown colored glass on either side .
2. Hb pipette which is marked up to 20 mm^3 (0.02ml blood) .
3. Tube with markings of Hb on one side .
4. Glass rod.
5. Dropper .

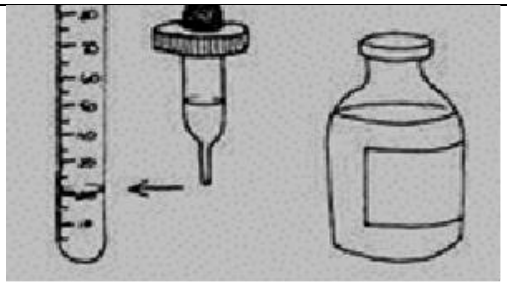
C. N/10 HCl .

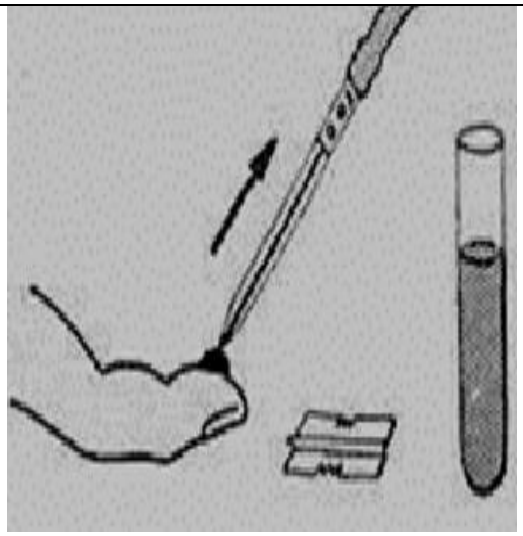
D. Distilled water.



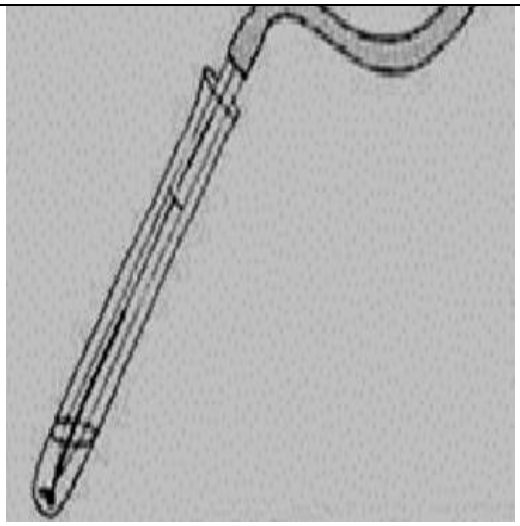
Pic : Hemocytometer .

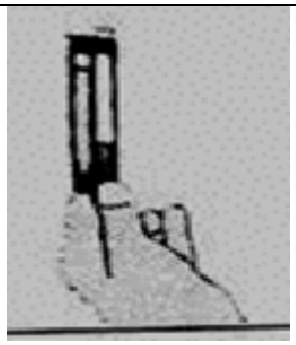
Procedure:

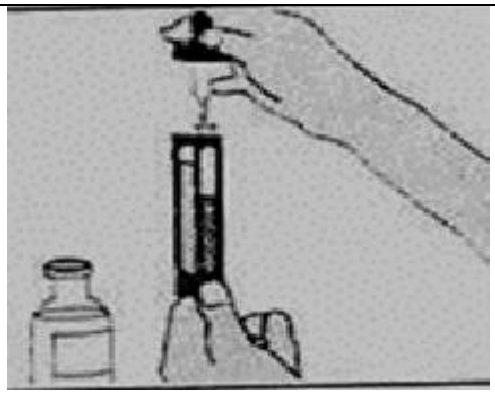
1. Fill the graduated tube to the 20 mark (or the mark 3 g / 100 ml) with 0.1 mol /l HCL 20 مع 0.1 HCL	

2. draw venous or capillary blood to the 0.02 ml mark of the Sahli pipette. Do not allow air bubbles to enter .With venous blood ensure that it is well mixed by inverting the bottle containing it and the anticoagulant repeatedly for about 1 minute immediately before pipetting it . (Do not take the first drop of blood from the finger.)	

3. Wipe the outside of the pipette with cotton.

4. Blow the blood from the pipette into the graduated tube of the acid solution. Mix gently by glass rod and leave it for 10 min.	

5. Place the graduated tube in the haemoglobinometer Stand facing a window. Compare the colour of the tube containing diluted blood with the colour of the standard tube.	

6. If the color is darker than that of the standard tube , continue to dilute by adding 0.1 mol/l HCL drop by drop. Stir with the glass rod after adding each drop. Remove the rod and compare the colour of the 2 tubes. Stop when the colours match . Distilled water can also be used at this step instead of o.1 mol/l HCL to continue the dilution of the blood.	

7. Remove the stirrer and take the reading directly by noting the height of the diluted acid hemat.

Normal value:

man		13.0 – 18.0
woman		11.5 – 16.5
Children	Infant	14.0 – 19.5
	3 months	9.5 – 13.5
	1 year	10.5 – 13.5
	3 – 6 year	12.0 – 14.0
	10 –12 year	11.5 – 14.5

Clinical significance:

Anemia: is a decrease in blood hemoglobin level . It could be due to:
1) Decreased production of hemoglobin as in:

a) Protein deficiency.
b) Iron deficiency.
c) Vitamin deficiency: Vit. B12, Folic acid, Vit. C.
d) Leukemia.

2)Increased destruction of hemoglobin as in:

a) Hemolytic anemia.
b) Heavy metal poisoning :Lead.
c) Infectious diseases :malaria.

Polycythemia: An increase in blood hemoglobin level. It could be due to: Congenital heart diseases.

Lecturer

Burhan Hadi