Estimation of Hemoglobin

Lab -3-

by

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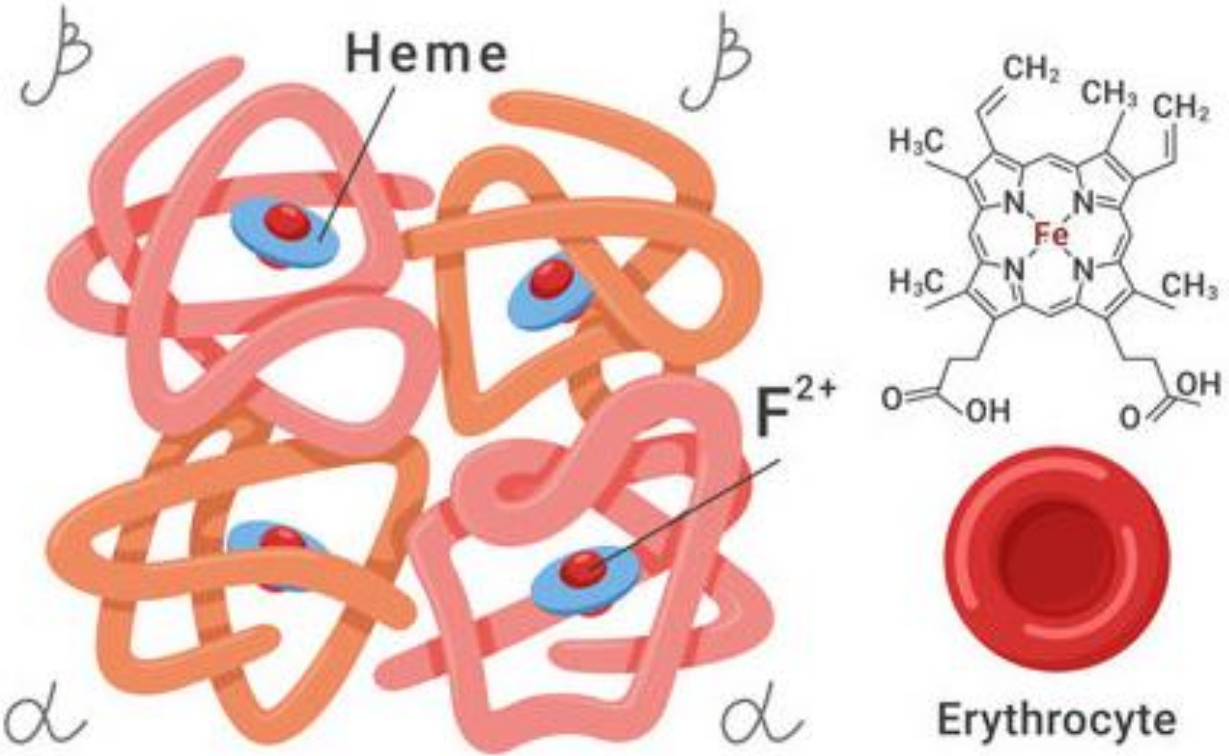
Introduction and principle

Hemoglobin is the iron-containing oxygen-transporter in the red blood cells of humans. Hemoglobin carries oxygen from the lungs to the tissues where it releases the oxygen to burn nutrients to provide energy to power the functions of the body, and collects the resultant carbon dioxide to bring it back to the lungs to be released to the atmosphere. Hemoglobin gives the blood its red color.

Hemoglobin is composed mainly of two parts; ***heme***, an oxygen-containing part and ***globin***, a protein.

The globin is normally composed of ***4 chains*** **of amino acids**; **two alpha (α) and two beta (β) chains.**

Structure Of Hemoglobin



Types of Normal Hemoglobin:

HbA: 2α 2β which is the normal adult hemoglobin, It is the predominant type of hemoglobin in adults.

HbF: 2α 2γ which is the main hemoglobin type in the fetus. HbF is replaced gradually by HbA soon after birth.

HbA_2:2α 2δ also normal in adult but it is less common**.**

Forms of Hemoglobin:

Oxyhemoglobin: when hemoglobin binds to oxygen, it is called oxyhemoglobin.

Carbaminohemoglobin: hemoglobin combined with CO_2, also called deoxyhemoglobin.

Carboxyhemoglobin: hemoglobin combined with CO.

Methemoglobin: a type of hemoglobin in which the iron in the heme group is in the Fe^{+3} (ferric) state, not the Fe^{+2} (ferrous) of normal hemoglobin.Methemoglobin cannot bind oxygen, unlike oxyhemoglobin.

Medical applications:

the normal range of Hb in healthy individuals is:

Male 13.5-18 g/dL

Female 11.5-16 g/dL

Newborn 21 g/dL

Medical Terminology:

Anemia: a condition of decreased level of hemoglobin **below 13.5 g/dL (males)** or **below 11.5 g/dL (females).** The hemoglobin level falls as red blood cells count decreases as in cases of **severe hemorrhage.**

Hemoglobin level is also decreased in case of **hemodilution, pregnancy** is an example.

Polycythemia: it literally means **increased red blood cell count, as RBC count raises**, the total level of hemoglobin is also **increased.**

Principle of experiment:

All the Hb is converted into acid hematine and the intensity of the color is measured by comparing it with a standard. This may be done visually.

The concentration of Hb is measured in **grams per deci-liter (g/dL).**

Materials and method:

Sahli’s hemoglobinometer consists of

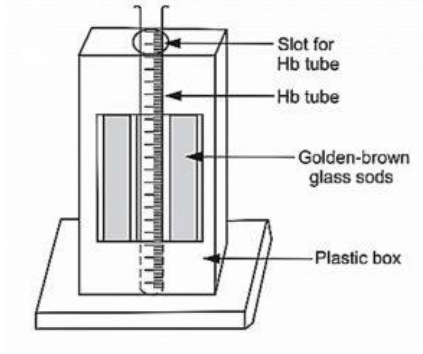
1. Pipette marked to contain 20 micro liters of blood.

2. Graduated tube

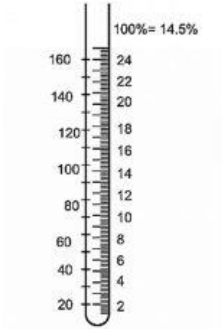
3. Distilled water (D.W.)

4. 0.1 normal HCL

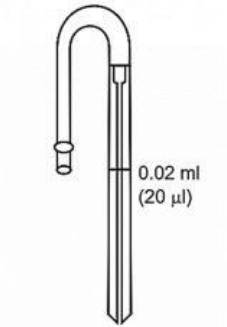
Sahli's Hemoglobinometer Set



Comparator



Hemoglobin tube



Hemoglobin pipette

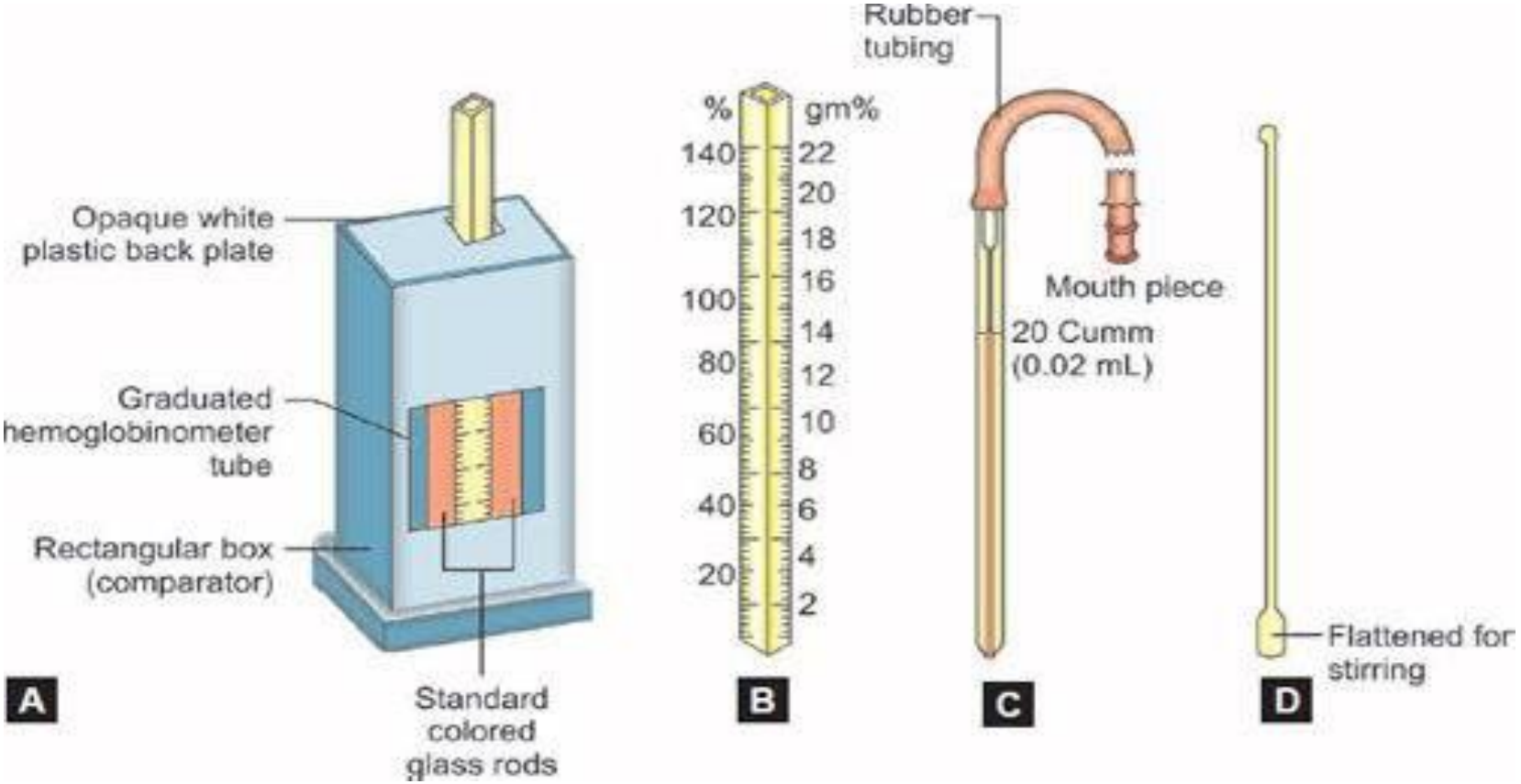
Stirrer

Procedure:

1. Fill the graduated tube to mark **(2 or 10)** with 0.1 normal HCl.
2. Draw blood by hemoglobin pipette to mark **20µL** (micro liters).
3. Dip the tip of the pipette in the graduated tube to blow the blood into the tube, mix content with a stirrer.
4. Place the tube in the hemoglobinometer for 10 minutes for complete reaction.
5. Add drop by drop D.W. until the color in the graduated tube is identical to the color of the standard.
6. Read the result in g/dL.

Sahli's hemoglobinometer (hemometer)


MARIENFELD
LABORATORY GLASSWARE
Haemometer
Made in Germany


Opaque white plastic back plate
Graduated hemoglobinometer tube
Rectangular box (comparator)
Standard colored glass rods
A
%
gm%
140
120
100
80
60
40
20
22
20
18
16
14
12
10
8
6
4
2
B
Rubber tubing
Mouth piece
20 Cumm (0.02 mL)
C
Flattened for stirring
D