INDICATIONS FOR HEMOGLOBIN ESTIMATION

1. To determine presence and severity of anemia: Anemia refers to low hemoglobin concentration

   or oxygen-carrying capacity of blood.

   Clinical signs of anemia (pallor of skin, conjunctival vessels, or

   mucous membranes are unreliable for diagnosis of anemia.

2. Screening for polycythemia: Polycythemia refers to

   increased hemoglobin level above the normal range.

   It may be primary, secondary, or relative .

3. To assess response to specific therapy in anemia.

4. Estimation of red cell indices (along with packed cell

   volume and red cell count) i.e. mean cell hemoglobin and mean cell hemoglobin concentration.

5. Selection of blood donors.

Sahli's Acid Hematin Method :

Principle: Blood is mixed with an acid solution so that hemoglobin is converted to brown-colored acid hematin. This is then diluted with water till the brown color matches that of the brown glass standard. The hemoglobin value is read directly from the scale.

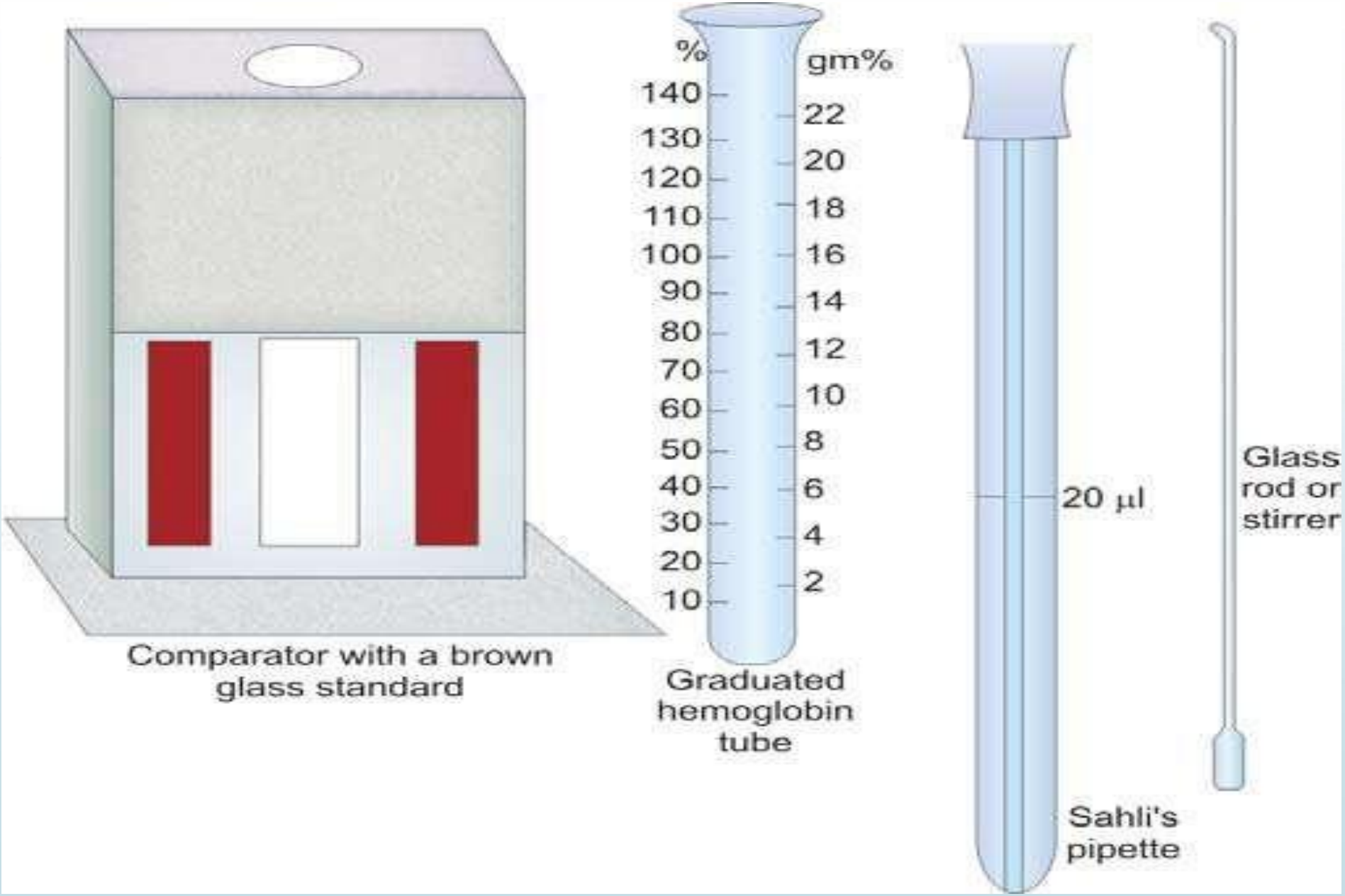
Equipment :

1. Sahli's hemoglobinometer: This consists of Sahli's graduated hemoglobin tube (marked in grams and percent) and a comparator with a brown glass standard.
2. 2. Sahli's pipette or hemoglobin pipette (marked at 20 µl or 0.02 ml).
3. Small glass rod (stirrer).
4. Dropping pipette.

Reagents :

1. N/10 hydrochloric acid
2. Distilled water

Specimen: EDTA-anticoagulated venous blood or blood obtained by skin puncture.


%
gm%
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20
18
16
14
12
10
8
6
4
2
20 µl
Glass rod or stirrer
Sahli's pipette
Comparator with a brown glass standard
Graduated hemoglobin tube

Method :

1.Place N/10 hydrochloric acid into Sahli's graduated hemoglobin tube up to the mark of 2 grams.

2.Take blood sample in Sahli's pipette exactly up to 20 µl mark. Blood adhering to the exterior of the pipette is wiped away using absorbent paper or gauze.

3.Add blood sample to the acid solution, mix with a glass stirrer, and allow to stand for 10 minutes.

4. Add distilled water drop by drop till the color of the solution matches that of the glass standard.

5.Take the reading of the lower meniscus from the graduated tube in grams

Disadvantages :

- About 95% color of acid hematin is attained at the end of 10 minutes. For maximum

  color development, much longer time (1 hour) is required.
- Perfect matching with the brown glass standard is not possible.
- Carboxyhemoglobin, methemoglobin, and sulfhemoglobin are not converted to

  acid hematin. HbF is also not converted to acid hematin and therefore this method

  is not suitable in small infants.
- Development of color is slow and acid hematin solution is not stable.
- Source of light (daylight or artificial) will influence

  the visual comparison of colors.
- Personal error in matching brown glass standard with test solution is 10%.
- Color of brown glass standard fades with time.

REFERENCE RANGES (WORLD HEALTH ORGANIZATION):

- Adult males: 13.0 - 17.0 gm/dl.
- Adult females (non-pregnant): 12.0 – 15.0 gm/dl.
- Adult females (pregnant): 11.0 - 14.0 gm/dl.
- Children, 6-12 years: 11.5 - 15.5 gm/dl.
- Children, 6 months to 6 years: 11.0 – 14.0 gm/dl.
- Children, 2 – 6 months: 9.5 – 14.0 gm/dl.
- At birth (full term): 13.6 – 19.6 gm/dl.

CRITICAL VALUES :

- < 7 gm/dl (severe anemia)
- > 20 gm/dl (hyperviscosity)

Other methods :

1. ***Colorimetric methods***: In these methods, color comparison is made between the standard and the test sample, either visually or by colorimetric methods.
   - Visual methods: Tallqvist chart, Sahli's acid hematin method, and WHO hemoglobin color scale.
   - Photoelectric methods: Cyanmethemoglobin (hemiglobincyanide) method, oxyhemoglobin method, and alkaline hematin method.

2.Gasometric method .

3.Chemical method.

4.Specific gravity method .