جامعة المستقبل
كلية التقنيات الصحية والطبية-قسم التخدير

Physiology Practical
Lecture: (5)
ESTIMATION OF HEMOGLOBIN
by
Sahli's/acid hematin Method

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General Objective of the Lecture:

To provide a comprehensive understanding of the hemoglobin estimation method using the Sahli's/acid hematin technique.

Behavioral Objectives:

1. Identify the fundamental principles of the Sahli's/acid hematin method for hemoglobin estimation.
2. Explain the process of converting hemoglobin to acid hematin using HCl.
3. Execute the steps for hemoglobin estimation using the required equipment and materials.
4. Evaluate the accuracy of the obtained results by comparing them with the normal hemoglobin values.

What is hemoglobin, and what is its primary function in the body?

Sahli's/acid hematin Method

Principle: Blood is mixed with N/10 HCl resulting in the conversion of Hb to acid hematin which is brown in color. The solution is diluted till it's color matches with the brown colored glass of the comparator box. The concentration of Hb is read directly.

- Hemoglobin + (0.1 N) HCl → Acid hematin (brown colour)

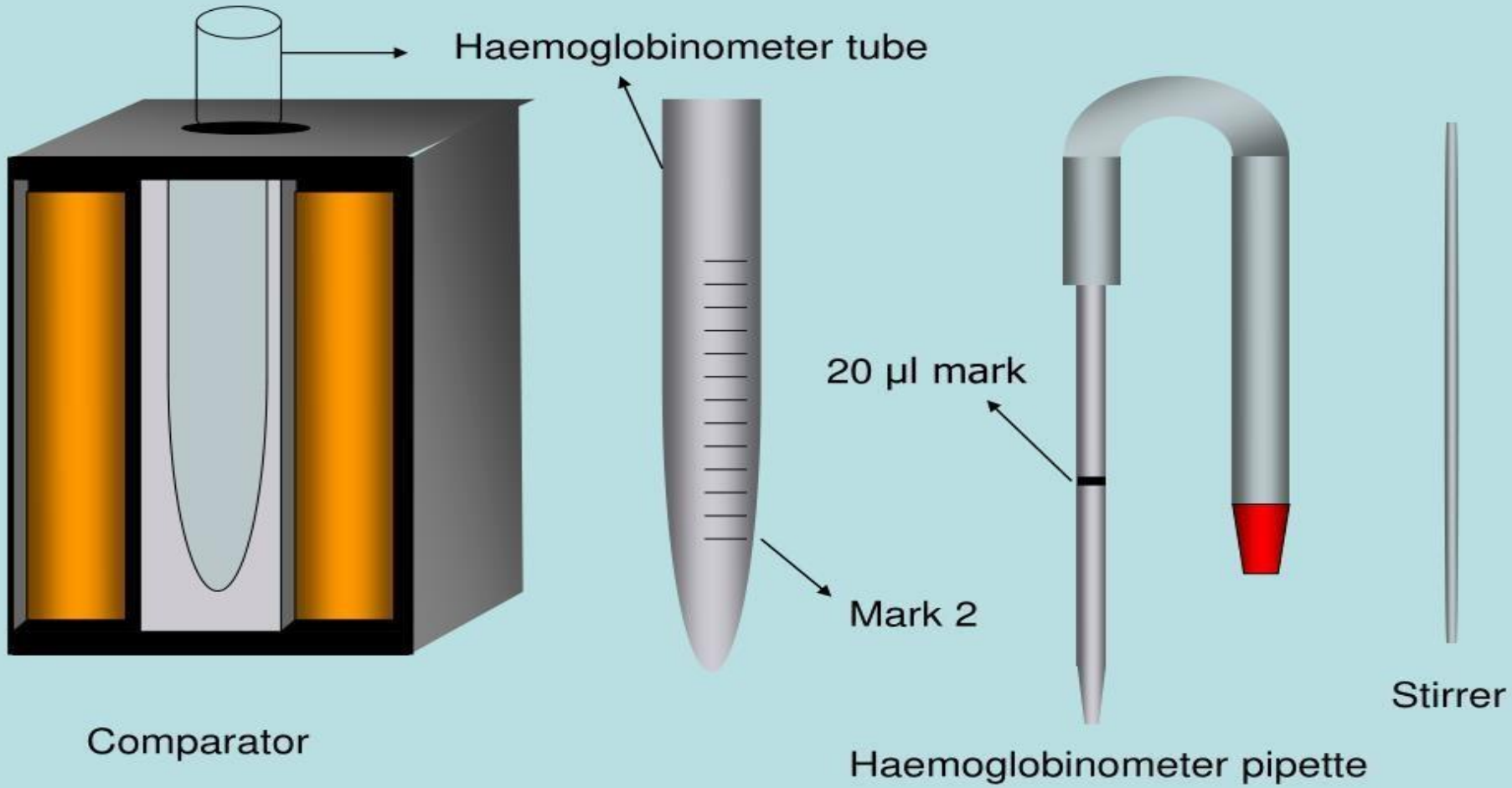
- The brown color of compound is matched against a brown glass standard in a comparator.

Equipment required

- **Hemocytometer** which consists of:

1. comparator box which has brown colored glass on either side
2. Hb pipette which is marked up to 20mm3(0.02ml blood)
3. Tube with markings of Hb on one side
4. glass rod
5. dropper

SAHLI'S HAEMOGLOBINOMETER



Reagents

- N/10 HCl
- Distilled water

Sample:

- Venous blood collected in EDTA as described earlier

Procedure

1. Placed 0.1(N)HCl acid in the Sahli's tube up to the lowest mark 20% by using a Pasteur pipette
2. Pipette 0.02 ml of blood in a Hb-pipette and added with the 0.1(N)HCl acid present in the Sahli's tube. Mixed well and wait for 10 minutes.
3. Diluted the solution with distilled water by adding few drops at a time carefully and diluted

• the solution, until the colour matches with the glass comparator present in the haemometer.

4. The colour matching should be done only against natural day light. The level of the fluid is noted at its lower meniscus and the reading corresponding to this level on the scale is recorded in gm/dl.

Group Activity:

Form small groups to discuss the advantages and disadvantages of the Sahli's/acid hematin method, and propose suggestions for improving the accuracy of the results.

Advantages

- Easy to perform
- Quick
- Inexpensive
- Can be used as a bedside procedure
- Does not require technical expertise

Disadvantages

- Less accurate.
- All hemoglobins (oxyhemoglobin, sulphemoglobin) are not converted to acid hematin and hence the value of Hb obtained is less than the actual value.
- The color of acid hematin develops slowly.
- Color of acid hematin fades with time and dilution must be done exactly after 10 min when the color development is maximum
- Individual variation in matching of color is seen.

Hemoglobin Interpretation

A. Increased values: Physiological

- High attitude.
- Young age.

Pathological

- Dehydration.

B. Decreased values :

Physiological

- Fluid therapy.

Pathological

- Anemia.
- Hemorrhage.
- Blood parasites.
- Malignant tumors.

Thank You!