



Store at: +2+8°C.

Presentation:

Cod. SU020 CONT: R 1 x 50 mL.

Cod. SU021 CONT: R 4 x 5 mL.

Procedure

Hemoglobin g/dL = (A) Sample x 37.7

Quantitative determination of Hemoglobin.

Only for in vitro use in clinical laboratory (IVD)

TEST SUMMARY

Hemoglobin is oxidized by potassium ferricyanide into methemoglobin, which is converted into cyanomethemoglobin, by potassium cyanide.

The intensity of the color formed is proportional to the Hemoglobin concentration in the sample^{1,2}.

REAGENTS COMPOSITION

| | | |
|---------------------------|---|--|
| Hemoglobin 50x | Potassium ferricyanide Potassium cyanide Dihydrogen potassium phosphate | 0.60 mmol/L 0.90 mmol/L 2 mmol/L |
| Hemoglobin CAL (Optional) | Hemoglobin Standard Animal origin | 15 g/dL |

PRECAUTIONS

HEMOGLOBIN Ref: SU020 / SU021

Harmful (Xn): R20/21/22: Harmful by inhalation, in contact with skin and if swallowed. S7: Keep container tightly closed. S28.1: After contact with skin, wash immediately with plenty of water. S45: In case of accident or if you feel unwell, seek medical advice immediately.

Cyanide (poison): The amount of cyanide in the Reagent Concentrate (50x) is appreciably less than the minimum lethal dose for an adult. Gaseous hydrogen cyanide will be released on contact with acids.

REAGENT PREPARATION AND STABILITY

Working reagent (WR):

- For 5 mL 4.9 mL of distilled water + 2 drops of Reagent
- For 250 mL 245 mL of distilled water + 1 vial (5 mL) of Reagent

Mix well.

Stability: 30 days at 2-8°C, protected from the sunlight.

All the components of the kit are stable until the expiration date on the label when stored at 2-8°C, protected from light and contamination prevented during their use.

Do not use reagents over the expiration date.

Signs of Reagent deterioration:

- Presence of particles and turbidity.
- Blank absorbance (A) at 540 nm. ≥ 0.01

All the reagents of the kit are stable up to the end of the indicated month and year of expiry. Store tightly closed at 2-8°C. Do not use reagents over the expiration date.

SPECIMEN

Venous or capillary blood¹.

Use anticoagulants like EDTA, heparin or oxalate.

Stability of the sample: 1 week at 2-8°C.

MATERIAL REQUIRED BUT NOT PROVIDED

- Spectrophotometer or colorimeter measuring at 540 nm.
- Matched cuvettes 1.0 cm. light path.

General laboratory equipment.

TEST PROCEDURE

1. Assay conditions:
Wavelength: 540 nm
Cuvette: 1 cm. light path
Temperature 15-25°C
2. Adjust the instrument to zero with distilled water.
3. Pipette:

| | | | |
|-----------------------|-------|----------|--------|
| | Blank | Standard | Sample |
| WR (mL.) | 5.0 | 5.0 | 5.0 |
| Hemoglobin Cal. (µL.) | -- | 20 | -- |
| Sample (µL.) | -- | -- | 20 |

4. Mix and incubate for 3 min. at room temperature (15-25°C).
5. Read the absorbance (A) of the samples and calibrator, against the Blank. The colour is stable for at least 48 hours.

CALCULATIONS

- With factor²:

- With calibrator:

$$\text{Hemoglobin g/dL} = \frac{(A)\text{Sample}}{(A)\text{Standard}} \times 15 \text{ (Standard conc.)}$$

QUALITY CONTROL

Each laboratory should establish its own Quality Control scheme and corrective actions.

REFERENCE VALUES

| | |
|-------|--|
| Men | 14 - 18 g/dL \cong 8.7 - 11.2 mmol/L |
| Women | 12 - 16 g/dL \cong 7.5 - 9.9 mmol/L |

(These values are for orientation purpose).

It is suggested that each laboratory establish its own reference range

CLINICAL SIGNIFICANCE

The Hemoglobin is a protein that contains iron and that the red color to the blood. The Hemoglobin is in red globules and it is the one in charge of oxygen transport by the blood from the lungs to tissues.

When the level of Hemoglobin appears underneath the normal levels is describing an anemia that can be of different origins: primary anemia, cancer, pregnancy, renal diseases, and hemorrhages.

If the Hemoglobin levels appear high it can be due to: cardiopathies, dehydration and stays in places of much altitude^{1,5,6}.

Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

REAGENT PERFORMANCE

- **Measuring Range:**

From detection limit of 0.1 g/dL. to linearity limit of 18 g/dL., under the described assay conditions.

If results obtained were greater than linearity limit, dilute the sample 1/2 with NaCl 9 g/L. and multiply result by 2.

- **Precision:**

| | Intra-assay n= 20 | | Inter-assay n= 20 | |
|-------------|-------------------|------|-------------------|------|
| Mean (g/dL) | 8.00 | 15.2 | 7.81 | 15.1 |
| SD | 0.29 | 0.33 | 0.19 | 0.26 |
| CV (%9) | 3.59 | 2.19 | 2.51 | 1.74 |

- **Sensitivity:**

1 g/dL. = 0.027A

- **Accuracy:**

Results obtained GPL reagents did not show systematic differences when compared with other commercial reagents.

The results of the performance characteristics depend on the analyzer used.

INTERFERING SUBSTANCES

- A list of drugs and other substances that could interfere has been reported by Young et. al^{5,6}.

NOTES

1. Use clean disposable pipette tips for its dispensation.

BIBLIOGRAPHY

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