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Liquid Reagents – ready to use

Hemoglobin Total

Cyanmethemoglobin

Single Reagent

Diagnostic Reagent for quantitative in vitro determination of Hemoglobin in human whole blood on photometric systems.

REF	Kit Size	Configuration
Y04704B	**1 x 1 L**	Single Reagent
Y04701	**5 x 100 mL**	Single Reagent
Y04702	**5 x 50 mL**	Single Reagent
Additionally offered:		
Y04705SV	1 x 2 mL	Hemoglobin Calibrator
Y04706	6 x 2 mL	Hemoglobin Control Set

TEST PARAMETERS

Method:	Colorimetric, endpoint, increasing reaction, Cyanmethemoglobin
Wavelength:	540 nm, 520 – 560 nm
Temperature:	Room temperature
Sample:	Whole blood with EDTA, oxalate, citrate or heparin as anticoagulants
Linearity:	up to 20 g/dL

SUMMARY

Previous methods used for the determination of blood haemoglobin were based on estimations of oxygen or carbon monoxide capacity or iron content. Of all methods, only the cyanmethemoglobin has gained popular acceptance.
The original cyanmethemoglobin technique was proposed by Stadie in 1920 [1]. This method used separate alkaline ferricyanide and cyanide reagents. A single reagent was introduced by Drabkin and Austin [2] in 1935. In 1958 the National Research Council (NRC) recommended adoption of the cyanmethemoglobin procedure based on field trials conducted by the Army Medical Department [3,4]. In 1966 the International Committee on Standardization in Hematology approved the proposal that all clinical laboratories should adopt this method exclusively [5].

TEST PRINCIPLE

In an alkaline medium, potassium ferricyanide oxidizes hemoglobin and its derivatives to methemoglobin. Subsequent reaction with potassium cyanide produces the more stable cyanmethemoglobin which has a maximum absorbance at 540 nm [6]. Color intensity is proportional to total hemoglobin concentration.
This procedure measures hemoglobin and its derivatives except sulfhemoglobin.

REAGENT COMPOSITION

COMPONENTS	CONCENTRATION
Potassium ferricyanide	0.6 mmol/L
Potassium cyanide	0.77 mmol/L
Buffers, stabilizers	

REAGENT PREPARATION

The reagent provided is ready to use.

REAGENT STABILITY AND STORAGE

Conditions:	protect from light close immediately after use
Storage:	at 15 – 25 °C
Stability:	up to the expiration date

Do not use reagent if it has become a different color than yellow or if it has become cloudy.

SAMPLE STABILITY AND STORAGE

Stability:	15 – 25 °C	1 week

Discard contaminated specimens.

MATERIALS REQUIRED BUT NOT PROVIDED

General laboratory equipment

MANUAL TEST PROCEDURE

Pipette into test tubes	Blank	Calibrator	Sample
Reagent	2000 µL	2000 µL	2000 µL
Sample	-	-	10 µL
Calibrator	-	10 µL	-
Mix, incubate for 3 min. at room temperature and read absorbance against reagent blank within 1 hour.			

CALCULATION

$$\text{Hemoglobin (g/dL)} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Calibr.}} \times \text{Conc. of Cal. (g/dL)}$$

REFERENCE RANGE [g/dL]* [11,7]

Adult Males:	13.0 – 18.0
Adult Females:	11.0 – 16.0
Children:	10.0 – 14.0
Newborns:	14.0 – 23.0

* Factors such as age, race, exercise, season and altitude are reported to influence the values of normal ranges. It is recommended that each laboratory establishes its own normal range.

PERFORMANCE CHARACTERISTICS

LINEARITY

The assay is linear to 20.0 g/dL.
Samples with hemoglobin concentrations higher than 20.0 g/dL must be re-run using one-half the sample volume. Multiply final results by two.

PRECISION

Assays (n=25) of haemoglobin control material yielded a coefficient of variation of 1.1% at 8.9 g/dL and 1.4% at 12.6 g/dL.

SPECIFICITY/INTERFERENCES

1. Substances that cause turbidity will falsely elevate the hemoglobin value. These include lipids [7], abnormal plasma proteins (macroglobulinemia) [8] or erythrocyte stroma [9].
2. A review by young et al. [10] reveals the numerous drugs that exert an in vitro effect to decrease blood hemoglobin values.

METHOD COMPARISON

Studies conducted against a similar procedure yielded a coefficient of correlation of 0.992 with a regression equation of y = 0.985 x + 0.098 on samples with values from 8.7 to 18.2 g/dL (n=27).

CALIBRATION

The assay requires the use of a hemoglobin calibrator.
We recommend the Dialab **Hemoglobin Calibrator**.

QUALITY CONTROL

All controls with hemoglobin values determined by this method can be used.
We recommend the Dialab **Hemoglobin Control Set**.

AUTOMATION

This assay is intended for manual use.

WARNINGS AND PRECAUTIONS

1. The reagent contains cyanide. Poison – may be fatal if swallowed. Do not pipette by mouth.
2. Do not mix with acids. Discard by flushing with large volumes of water.
3. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents.

4. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
5. For professional use only!

WASTE MANAGEMENT

Please refer to local legal requirements.

REFERENCES

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