

HEMOGLOBIN A1C

Turbidimetry

The application parameters comprised here constitute a guide to facilitate the validation of our reagents by the instrument. It is advisable to validate the use when there is any change in software or reagent versions.

Instrument: **HITACHI 717**

Samples

Capillary or venous blood collected by standard procedures and with heparin or EDTA as anticoagulants .
 HbA1C in blood is stable 3 days at 15-25°C, 7 days at 2-8°C and 6 months at -20°C. Freeze once only.

Hemolysate preparation

The calibrators do not require pretreatment.

1. Bring the reagent A to room temperature.
2. Pipette into a test tube:

Blood	10 µL
Reagent (A)	1000 µL

3. Shake thoroughly. Avoid the formation of foam. The hemolysate can be used after the solution has changed color from red to brownish-green (approximately 3 minutes).

The hemolysate is stable 4 hours at 15-25°C, 24 hours at 2-8°C and 6 months at -20°C. Freeze once only.

Reagent preparation

Reagents (A), (B), (C) and (D) are provided ready to use.
 HbA1C Standards (S1-S4): Reconstitute with 2.0 mL of distilled water. Stable for 8 hours at 15-25 °C, 2 days at 2-8°C and 3 months at -20°C. Freeze once only.

Hb Reagent 1: Reagent B
 HbA1c Reagent 1: Reagent C Reagent 2: Reagent D

Instrument settings

TEST	HBA1C	HB
ASSAY CODE	2 POINT - 24 - 50	1 POINT - 24 - 0
SAMPLE VOLUME (µL)	10 - 5	20 - 10
R1 VOLUME (µL)	250 - 20 - 0	200 - 20 - 0
R2 VOLUME (µL)	50 - 20 - 0	0 - 20 - 0
WAVELENGTH (nm)	700 - 340	660 - 570
CALIB. METHOD	3 - 1 - 5	1 - 0 - 0
STD 1 CONC. POS.	0.00 - 1	0.0 - 1
STD 2 CONC. POS.	(*) - 2	(*) - 5
STD 3 CONC. POS.	(*) - 3	0 - 0
STD 4 CONC. POS.	(*) - 4	0 - 0
STD 5 CONC. POS.	(*) - 5	0 - 0
STD 6 CONC. POS.	0 - 0	0 - 0
SD LIMIT	300	0.1
DUPLICATE LIMIT	400	300
SENSITIVITY LIMIT	2000	0
ABS. LIMIT	0 - 0	0 - 0
PROZONE LIMIT	0 - 0	0 - 0
EXPECTED VALUE	... - - ...
PANIC VALUE	0.05 - 2.5	1.1 - 40
INSTRUMENT FACTOR	1.0	1.0

Blank: Make reagent blank with sodium chloride 154 mmol/L.
 Calibrators 2-5: Standards S1-S4 Calibrator: Standard S4

CALCULATION

$$\% \text{ HbA1C - IFCC} = \frac{\text{HbA1C (g/dL)}}{\text{Hb (g/dL)}} \times 100$$

The HbA_{1c} percentage in the sample is calculated using the following general formula. The values are traceable to IFCC Reference Method:

The traceable values to Reference Method as described by the US National Glycohemoglobin Standardization Program (NGSP) are calculated using the following general formula:

$$\% \text{HbA1C-NGSP} = 0.915 \times \% \text{HbA1C-IFCC} + 2.15$$