

# 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: **MONARCH**

### 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.**

### 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

Hb

Identification parameters		Data acquisition parameters	
Test code	...	Analysis type	MIX
Test name	Hb		RUN
Test mnemonic	HB	Temperature	37°C
Optical mode	ABSORBANCE	Delay time	300 sec
Response algorithm	FINAL-INITIAL	Interval time	10 sec
Result algorithm	LINEAR	N° of data pts.	2
<b>Loading parameters</b>		Filter 1	690 nm
Loading type	LOAD	Filter 2	550 nm
	ANALYZE	Monochromator 1	690 nm
Reagent blank	ON	Monochromator 2	550 nm
Reference type	DILUENT	Compatibility	10
Calibrator type	TEST-SPECIFIC (...)	<b>Data integrity parameters</b>	
Sample volume	30 µL	Integrity tests	SLOPE
Sample diluent	0 µL		NORMAL RANGE
Reagent diluent	10 µL		MINIMUM ABS/INT
1 <sup>st</sup> reagent (R1)	170 µL		MAXIMUM ABS/INT
2 <sup>nd</sup> reagent (R2)	0 µL	<b>Integrity parameters</b>	
1 <sup>st</sup> reagent bar code	...	Slope	POSITIVE
2 <sup>nd</sup> reagent bar code	...	Lower limit	1.1 g/dL
<b>Data fit parameters</b>		Upper limit	40.0 g/dL
Calibrator	0.00	Minimum abs/int	0.0
	(*) S4	Maximum abs/int	0.1
Correction mode	NONE	Blank: Make reagent blank with sodium chloride 154 mmol/L.	
Units	g/dL	(...) Information entered by the user	
N° of dec. Places	1	(*) Enter the Standard assigned value	

## Hb A1c

<p><b>Identification parameters</b></p> <p>Test code ...</p> <p>Test name <b>Hb A1c</b></p> <p>Test mnemonic <b>Hb A1c</b></p> <p>Optical mode <b>ABSORBANCE</b></p> <p>Response algorithm <b>FINAL-INITIAL</b></p> <p>Result algorithm <b>NON LINEAR INTERPOLATION</b></p> <p><b>Loading parameters</b></p> <p>Loading type <b>LOAD</b></p> <p style="padding-left: 20px;"><b>ANALYZE</b></p> <p>Reagent blank <b>ON</b></p> <p>Reference type <b>DILUENT</b></p> <p>Calibrator type <b>TEST-SPECIFIC (...)</b></p> <p>Sample volume <b>8 µL</b></p> <p>Sample diluent <b>0 µL</b></p> <p>Reagent diluent <b>10 µL</b></p> <p>1<sup>st</sup> reagent (R1) <b>200 µL</b></p> <p>2<sup>nd</sup> reagent (R2) <b>40 µL</b></p> <p>1<sup>st</sup> reagent bar code ...</p> <p>2<sup>nd</sup> reagent bar code ...</p> <p><b>Data fit parameters</b></p> <p>Calibrator <b>0.00</b></p> <p style="padding-left: 20px;">(*) S4</p> <p style="padding-left: 20px;">(*) S3</p> <p style="padding-left: 20px;">(*) S2</p> <p style="padding-left: 20px;">(*) S1</p> <p>Correction mode <b>NONE</b></p> <p>Units <b>g/dL</b></p> <p>N° of dec. Places <b>2</b></p>	<p><b>Data acquisition parameters</b></p> <p>Analysis type <b>MIX</b></p> <p style="padding-left: 20px;"><b>RUN</b></p> <p>Temperature <b>37°C</b></p> <p>Delay time <b>300 sec</b></p> <p>Interval time <b>300 sec</b></p> <p>N° of data pts. <b>2</b></p> <p>Filter 1 <b>690 nm</b></p> <p>Filter 2 <b>340 nm</b></p> <p>Monochromator 1 <b>690 nm</b></p> <p>Monochromator 2 <b>340 nm</b></p> <p>Compatibility <b>10</b></p> <p><b>Data integrity parameters</b></p> <p>Integrity tests <b>SLOPE</b></p> <p style="padding-left: 20px;"><b>NORMAL RANGE</b></p> <p style="padding-left: 20px;"><b>MINIMUM ABS/INT</b></p> <p style="padding-left: 20px;"><b>MAXIMUM ABS/INT</b></p> <p>Integrity parameters</p> <p style="padding-left: 20px;">Slope <b>POSITIVE</b></p> <p style="padding-left: 20px;">Lower limit <b>0 g/dL</b></p> <p style="padding-left: 20px;">Upper limit <b>40 g/dL</b></p> <p style="padding-left: 20px;">Minimum abs/int <b>0.0</b></p> <p style="padding-left: 20px;">Maximum abs/int <b>0.1</b></p> <p>Blank: Make reagent blank with sodium chloride 154 mmol/L.</p> <p>(...) Information entered by the user</p> <p>(*) Enter the Standard assigned value</p>
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Version 0711

### CALCULATION

The HbA<sub>1c</sub> percentage in the sample is calculated using the following general formula:

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

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:

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

### HOW TO PROGRAM THE CALCULATED TESTS?

The Derived Calculations are defined and established by the user. Various combinations of tests and functions can be set up.

To define a new Derived calculation proceed as follows:

1. Press the desired number followed by the Enter key
2. Enter the user name or initials (to identify the test) followed by the Enter key
3. A table appears showing the code letters to use for the various functions:  
Test(A) , Const(B), +(C), -(D), \*(E), /(F), ( (G), ) (H)
4. Press the A key to place the cursor into the test box, then press the desired chemistry key.
5. Continue in the same manner by pressing the letter code for each desired function until the calculation is completed.
6. Terminate the calculation by pressing the Enter key.