



# HbA1c DIRECT

1 x 15 / 1 x 5 / 1 x 32 / 4 x 0.5 mL  
51835001

2 x 15 / 2 x 5 / 1 x 63 / 4 x 0.5 mL  
51835002

### INTENDED USE

This reagent is intended for *in vitro* quantitative determination of HbA1c in human blood.

- Latex enhanced Immunoturbidimetry
- Ready to use liquid stable reagents
- Multipoint calibration
- Direct result (% HbA1c) from analyzer
- No total Hb determination required

### CLINICAL SIGNIFICANCE

HbA1c is a glycated form of haemoglobin formed by the attachment of glucose residues in the blood to the hemoglobin molecules. In the diabetic population where blood glucose levels are abnormally elevated the level of HbA1c also increases. The level of HbA1c is proportional to the level of glucose in the blood and has been widely accepted as an indicator of the mean blood glucose concentration in the preceding 6-8 weeks. It is therefore a long-term indicator of diabetic control. For routine use HbA1c levels should be monitored every 3-4 months. However in gestational diabetes and after a change in therapy it may be useful to measure HbA1c more frequently at 2-4 week intervals.

### PRINCIPLE

The principle of the test is latex agglutination method that measures the ratio of hemoglobin A1c that occupy in a total hemoglobin in the whole blood. The sample (hemolysis sample) is added to the unsensitized latex particles, and the surfaces of the latex adsorb a total hemoglobin in the sample. Anti human HbA1c mouse monoclonal antibody complex agglutinates by anti-mouse IgG goat antibody. At this time, the amount of agglutination caused depends on the amount of HbA1c that adsorbs the surface of the latex, this agglutination is measured as a turbidity. The concentration of HbA1c (%) in the sample is determined by referring to the calibration curve obtained by the same test of diluted standard solutions.

### REAGENT COMPOSITION

<b>HbA1c Direct R1</b>	<b>1 x 15 mL</b>	<b>2 x 15 mL</b>
Latex		
<b>HbA1c Direct R2</b>	<b>1 x 5 mL</b>	<b>2x 5 mL</b>
Anti-human HbA1c mouse monoclonal antibody		
Anti-mouse IgG goat antibody		
<b>HbA1c R3</b>	<b>1 x 32 mL</b>	<b>1 x 63 mL</b>
Haemolysis Reagent		
<b>HbA1c DIRECT CALIBRATOR</b>	<b>4 x 0.5 mL</b>	<b>4 x 0.5 mL</b>
HbA1c 4 Level Calibrator (Lyophilized)		

### STORAGE AND STABILITY

The sealed reagents are stable up to the expiry date stated on the label, when stored at 2- 8°C, protected from light. Once opened the reagent is stable upto four weeks, if contamination is avoided. Recalibration is recommended after 30 days. **DO NOT FREEZE**  
**Calibrator : Reconstitute the calibrator with 0.5mL distilled water. It is stable for 30 days at 2-8°C . DO NOT FREEZE.**

### LINEARITY RANGE

The reagent is linear upto 13% (NGSP)

### REFERENCE RANGE

It is recommended that each laboratory establish its own reference values. The following value may be used as guide line.  
Reference normal value (NGSP): 4.6% -6.2%

### PRECAUTION

To avoid contamination, use clean laboratory wares. Use clean, dry disposable pipette tips for dispensing. Close reagent and standard bottles immediately after use. Avoid direct exposure of working reagent to light.

### SAMPLE

Whole blood, collected with EDTA

To determine HbA1c, a heamolysate must be prepared for each sample

1. Dispense 0.5 mL hemolysis reagent into a tube.
2. Add 10 µL of well-mixed whole blood and mix.
3. Allow to stand for 5 minutes or until complete lysis is evident.

Follow the same procedure with controls and calibrator.

### GENERAL SYSTEM PARAMETER

Mode of Reaction	FullyAuto
Slope of Reaction	Increasing
Wavelength	660 nm
Temperature	37°C
Calibrator Concentration	As on vial label
Linearity	13%
Blank	Reagent blank
Sample volume	7.5 µL
Reagent 1 Volume	180 µL
Reagent 2 Volume	60 µL
Cuvette	1 cm light path

### LABORATORY PROCEDURE FOR FULLY AUTO

	Blank	Calibrator	Sample/control
HbA1c R1	180 µL	180 µL	180 µL
Calibrator	-	7.5µL	-
Hemolysate(sample/control)	-	-	7.5µL

Mix & incubate for 5 min at 37°C.

	Blank	Calibrator	Sample/control
HbA1c R2	60 µL	60 µL	60 µL

Mix and incubate for 5 min at 37°C and read absorbance(A) at 660 nm.

### CALCULATION

Calibration curve

Calculate the ΔAbs of calibrators = Abs calibrator – Abs Blank. Plot the Δ Abs of each calibrator versus assigned concentration (HbA1c %) on a linear graph paper. HbA1c results according to NGSP for the samples and controls are determined using the prepared calibration curve.

Calculate

ΔAbs of sample ie abs Sample - abs Blank .

HbA1c % in the sample is calculated by interpolation of Abs of sample on the calibration curve. For calculation of results according to IFCC, use IFCC calibrator values (see calibrator insert), or use following equation.

NGSP = (0.915 x IFCC) + 2.15

### INTERFERENCES

No interference upto :

Ascorbic acid	50 mg/dL
Bilirubin	40mg/dL
Intra lipid	3000 mg/dL

It has been reported that results may be inconsistent in patients who have the following conditions: opiate addiction, lead poisoning, alcoholism, and ingestion of large doses of aspirin. Elevated HbF levels may lead to under estimation of HbA1c.

### BIBLIOGRAPHY

1. Engbeak, F., *et al.* Clin chem.35 p. 93-97 (1989)
2. American Diabetes Association : Clinical practice recommendations (position statement). Diabetes care 24 (suppl.1) S33-S55, (2001).
3. Tietz, N.W. Textbook of Clinical Chemistry, W.B. Saunders Company, p.794 - 795 (1999).

SYMBOLS USED ON THE LABELS: IN VITRO DIAGNOSTIC USE SEE PACKAGE INSERT FOR PROCEDURE LOT NUMBER MANUFACTURER'S ADDRESS MANUFACTURING DATE EXPIRY DATE TEMPERATURE LIMIT

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ADS/V.02/060114

ISO 9001 : 2008  
ISO 13485 : 2003