# HbA1c

**Glycated Hemoglobin A1c**

**Product information**

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|---|---|---|
| **24A1C01-UN** | **Meditest HbA1c** | **4x18 mL 2x12 mL** |
| **24A1C01-AU** | **Meditest HbA1c** | **4x18 mL 2x12 mL** |
| **24A1C01-AB** | **Meditest HbA1c** | **4x18 mL 2x12 mL** |
| **24A1C01-ER** | **Meditest HbA1c** | **4x18 mL 2x12 mL** |
| **24A1C01-AR** | **Meditest HbA1c** | **4x18 mL 2x12 mL** |

**Purpose**

In vitro test for the quantitative determination of glycated hemoglobin in human whole blood.

**Summary**

Hemoglobin A1c is continuously formed by the addition of glucose to the N-terminus of the hemoglobin beta chain. This non-enzymatic process reflects the average exposure of hemoglobin to glucose over a long period of time. Hemoglobin A1c in diabetic individuals has been shown to be 2-3 times higher than the levels found in normal patients. Many researchers have proposed the use of Hemoglobin A1c as an indicator of metabolic control of diabetic patients.

**Test principle**

This reagent uses antigen and antibody interaction to directly determine HbA1c in whole blood. Total hemoglobin and HbA1c have the same nonspecific rate of absorption into latex particles. When mouse antihuman HbA1c monoclonal antibody (R2) is added, the latex-HbA1c-mouse anti human HbA1c antibody complex is formed. Agglutination occurs when the goat anti-mouse IgG polyclonal antibody interacts with the monoclonal antibody. The amount of agglutination is proportional to the amount of HbA1c absorbed into the surface of the latex particles. The amount of agglutination is measured as absorbance. The HbA1c value is obtained from a calibration curve.

**Reagents - working solutions**

| | | |
|---|---|---|
| **R 1** Latex | Latex | <0.15% |
| **R 2** Antibody | Mouse anti-human HbA1c monoclonal antibody | <0.06 mg/mL |
| | Goat anti-mouse IgG polyclonal antibody | <0.09 mg/dL |

**Precautions warnings**

It is intended for in vitro diagnostic use by healthcare professionals. Follow the normal precautions necessary in handling all laboratory reagents.
Infectious or microbial waste:
Warning: handle waste as potentially biohazardous. Dispose of waste according to accepted laboratory instructions and procedures.
Environmental hazards: Follow all relevant local disposal regulations to determine that it has been disposed of safely. If requested, a safety data sheet can be provided to professional users.
Inhibit foam formation in all reagents and sample types (sample, calibrator and control).
If there is any damage on the package, do not use it Read the user manual carefully before use, do not use the expired assay kit Do not mix different lot reagents.
All samples should be considered epidemic material, please dispose of them in accordance with the laboratory working standard of infectious diseases.
Take the necessary protective measures to prevent users from becoming infected during operation.

**Use of reagents**

Ready to use.

**Storage and stability**

All components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8ºC, protected from light and contamination is avoided during their use.
Do not use reagents after the expiration date. Signs of reactive decay: Presence of particles and turbidity- empty absorbance (A) at 340 nm <1.00.

**Sample collection and preparation**

The test is formulated for use with human whole blood samples. Venous whole blood specimens collected with EDTA anticoagulant can be used. When stored in the refrigerator, it is recommended to use the samples within 7 days of collection. Prior to testing, whole blood samples should be mixed by slightly inverting them to resuspend the precipitated erythrocytes.
Use of automatic analyzer: Samples should be tested in stat mode (Emergency mode) to avoid precipitation.

**Required Materials (not included in the kit)**

1. Cat# 24A1C01-DC Meditest Diachem HbA1c Calibrator
2. Cat# 24A1C01-DQ Meditest Diacheck HbA1c Norm Control
3. Cat# 24A1C01-DQ Meditest Diacheck HbA1c Path Control
4. General laboratory equipment
5. Distilled or deionized water

**Working Procedure**

If you are using a spectrophotometer to perform this test, work with the following procedure. Ask your representative for the application data for fully automatic devices.

**Hemolysate Preparation**

1. Whole blood samples are taken to room temperature,
2. Blood samples are mixed so that the erythrocytes are mixed homogeneously,
3. Using a calibrated pipette, 1000 µL of Lyse solution is transferred to the sample beaker,
4. Transfer 40 µL of homogenized blood sample to the sample cup with Lyse added,
5. Hemolysate is thoroughly mixed, incubated at room temperature for 5 minutes,

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6. Hemolysate is ready to use for HbA1c

1.Test Conditions:
Wavelength: . 660 nm
Cuvette: . . . . . 1 cm light path
Temperature: .
2.Set the appliance to zero with distilled water.
3.Place the pipettes in a cuvette.

| | |
|---|---|
| R1 (µΛ) | 450 |
| Sample (µΛ) | 15 |

4.Stir and incubate for 2 minutes. Read absorbance after 30 seconds (A1)
5. Add on

| | |
|---|---|
| R2 (µL) | 150 |

6. Stir and read the absorbance (A2) 5 min after the addition of R2

**Calculation**

$$\text{HbA1c concentration (\%)} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ STD/CAL}} \times \text{Conc. STD}$$

ΔA= (A2-A1)

Conversion Formula: NGSP%= [0.09148 x (IFCC)] + 2.152

**Expected values**

4.5-6.5% (NGSP/DCCT)
26-48 mmol/mol (IFCC)
According to the data provided by the NGSP, HbA1c levels above 6.5% are suitable for the diagnosis of diabetes mellitus. Patients with an HbA1c between 39-46 mmol/mol (IFCC) or 5.7% - 6.4% HbA1c (NGSP) are at risk of developing diabetes. It is recommended that each laboratory establish its own normal range. The reference range was validated using the CLSI EP28-A3c protocol.

**Limitations**

- The linearity of the test is up to 15.5% HbA1c. Samples with values above 15.5 should not be diluted and retested. Instead, values should be reported as higher than 16% (>16%).
- It has been observed that alcoholism, high doses of acetyl salicylic acid, opiates, and lead poisoning may lead to inconsistency in patients
- The test is formulated for use with human whole blood specimens with EDTA
- High HbF levels may lead to inadequate evaluation of HbA1c and do not interfere with HbA1c determination by uremia immunoassay.

**Performance characteristics**

| Whole blood application | |
|---|---|
| n | 100 |
| Slope | 1.001 |
| Intercept | 0.027 |
| Correlation coefficient | 0.990 |
| Range of values | 4% - 15.5% HbA1c |

**Precision**

| | Intra-assay | | | Inter-assay | | |
|---|---|---|---|---|---|---|
| | Mean (%) | %CV | n | Mean (%) | %CV | n |
| Low | 5.46 | 1.45 | 80 | 5.46 | 2.81 | 80 |
| High | 10.1 | 1.73 | 80 | 10.1 | 2.72 | 80 |

* Sensitivity Studies data were validated using CLSI EP05-A3 protocol.
The results of the performance characteristics depend on the analyzer used.

**References**


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