Bioway Chemistry Reagent Series

Hemoglobin A1c 2parts Reagent Kit

Detection of Hemoglobin A1c in Human Blood on Chemistry Analyzers

Cat. No. R060K11 ***Hemoglobin A1c 2parts Reagent Kit***

SUMMARY OF TEST PROCEDURE

Figure 1: Procedure Diagram

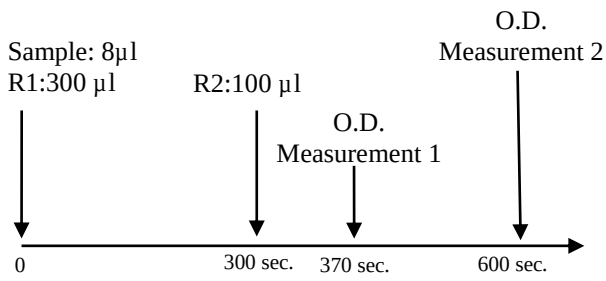

Table 1: Instrument Parameters*

Calibration method	5 point non-linear	Slope of reaction	increase
Wavelength	660nm	Sample volume	8 µl
Test method	2 point end	R1 volume	300 µl
Reaction temperature	37℃	R2 volume	100 µl

*Refer to Figure 1 and the package insert for detail

INTENDED USE

Bioway Chemistry Reagent Series **Hemoglobin A1c 2parts Reagent Kit** (the Kit) is a latex-enhanced immunoturbidimetric assay intended for *in vitro* quantitative detection of Hemoglobin A1c (HbA1c) in human blood on automated clinical chemistry analyzers.

SUMMARY AND EXPLANATION

Hemoglobin A1c is a subtype of hemoglobin A that is formed by a non-enzymatic process that adducts glucose to the N-terminal of the hemoglobin beta chain. This process reflects the average hemoglobin exposure to glucose over an extended period and provides clinical significance in monitoring the blood glucose level. Studies have shown HbA1c in diabetic patients to be 2-3 times the levels found in normal individuals. HbA1c can be used as an indicator of metabolic control of the diabetic.

TEST PRINCIPLES

The Kit utilizes the antibody-antigen reaction to directly measure the HbA1c level in whole blood. The first reaction, occurring after the sample is mixed with R1, consists of unspecified binding of total hemoglobin and HbA1c to the latex particles at the same rate. The second reaction occurs after the addition of R2 that contains mouse anti-human monoclonal antibody and goat anti-mouse IgG polyclonal antibody. Agglutination complexes will be formed from the interaction of the HbA1c bound to the latex particles with the respective antibodies. The agglutination can be measured as an absorbance which is proportional to the amount of HbA1c bound to the latex, and because the total hemoglobin and HbA1c bind to the latex at the same rate, the % HbA1c in total hemoglobin can be obtained from a calibration curve.

MATERIALS PROVIDED

Reagents:

R1	Latex 0.1%; Glycine buffer, pH 3.0, 15mmol/L
R2	goat anti-mouse IgG polyclonal antibody 0.8mg/dl; Mouse anti-human HbA1c monoclonal antibody 0.05 mg/ml; Glycine buffer, pH 3.0, 60mmol/L
Hemolysis reagent	H_2O, stabilizers

MATERIALS NEEDED BUT NOT PROVIDED

1. Pipette for 20 µl and 1 ml.
2. Test tube for applicable instrument
3. HbA1c calibrator set and control set (available for purchase)

INSTRUMENT

The Kit is applicable on most automated chemistry analyzers. Refer to specific instrument application for suggested settings.

STORAGE AND STABILITY

Store the reagents at 2-8℃. Avoid direct sunlight. The Kit is stable through the expiration date when stored properly. R1 and R2 reagents are stable for 1 month at 2-8℃ after opening.

PRECAUTIONS

1. The Kit is for *in vitro* diagnostic use only. Not for use in humans or animals.
2. The instructions must be followed to obtain accurate results.
3. Do not use the reagents beyond the expiration date.
4. Treat all specimens as infectious. Proper handling and disposal procedures of specimens and test materials should be strictly followed.

SPECIMEN COLLECTION AND HANDLING

The Test can be performed with human blood without special preparation of the patient. Follow standard laboratory procedures to collect specimens with EDTA.

Hemolysate preparation:

1. Mix 1 ml of Hemolysis Reagent with 20 µl of well mixed whole blood
2. Wait for 5 minutes or until complete lysis is evident before using the sample.
3. If immediate testing is not possible, hemolysates may be stored up to 10 days at 2-8℃.

TEST PROCEDURE (see Figure 1)

Calibration: 5 level calibrator set available for purchase. Recommend using Bioway calibrators for optimal results. Use multi-point non-linear calibration method.

Test procedure: see Figure 1 and Table 1 for instrument parameter setup. Refer to specific instrument application for suggested setting.

1. Add 8 µl of sample and 300 µl of R1, mix well and incubate at 37℃ for 300 seconds.
2. Add 100 µl of R2, mix well and incubate at 37℃.
3. Take optical density measurement OD 1 at 370 seconds.
4. Take optical density measurement OD 2 at 600 seconds.
5. Calculate ΔOD = OD 2 – OD 1

RESULT

The % HbA1c can be obtained by using the calculated ΔOD to find the corresponding % HbA1c on a calibration curve.

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EXPECTED VALUES

Less than 6% for non-diabetic person, less than 7% for glycemic control of a person with diabetes.
It is recommended for each laboratory to establish its own expected values

QUALITY CONTROL

Commercially available HbA1c controls may be used. It is recommended to use Bioway HbA1c Control set (available for purchase) for optimal results.
A control should be tested before each batch of tests to ensure the test is properly performed and all reagents and the instrument are functional as specified.

LIMITATIONS

1. The Kit is for *in vitro* use on automated chemistry analyzers only.
2. The Kit should not be used for the diagnosis of diabetes mellitus.
3. Results may be inconsistent in patients with the following conditions: opiate addiction, lead-poisoning, alcoholism, ingest large doses of aspirin.
4. Hemoglobin variants HbA2, HbC and HbS do not interference with the test. HbE has not been assessed.
5. Elevated levels of HbF may lead to underestimation of HbA1c.

PERFORMANCE CHARACTERISTICS

Linearity: 2% - 14% (R≥0.990)

Precision: Within Run: CV≤5%;
Run-to-Run: CV≤5%

Interference: no interference detected for: Bilirubin (0.5 g/L), ascorbic acid (0.5 g/L), triglycerides (20 g/L), carbamylated Hb (7.5 mmol/L), and acetylated Hb (5.0 mmol/L)

Reagent Blank Absorbance: at 660nm wavelength and 10 mm optical diameter, O.D. ≤ 1.00

REFERENCES

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Not Intended for Sale in the United States.

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