BIO TECHNOLOGY boditech

Diabetes ichroma™ HbA1c

INTENDED USE

ichroma™ HbA1c is a fluorescence Immunoassay (FIA) for the quantitative determination of HbA1c(Hemoglobin A1c) in human whole blood. It is useful as an aid in management and monitoring of the long-term glycemic status in patients with diabetes mellitus.

For *in vitro* diagnostic use only.

INTRODUCTION

Glycated protein is formed post-translationally through the slow, nonenzymatic reaction between glucose and amino groups on proteins. HbA1c is a clinically useful index of mean glycemia during the preceding 120 days, the average life span of erythrocytes. Carefully controlled studies have documented a close relationship between the concentrations of HbA1c and mean glycemia. HbA1c is considered as a more reliable parameter in monitoring glycemia over the glycemic reading with the conventional glucometer.

PRINCIPLE

The test uses a sandwich immunodetection method; the detector antibodies in buffer bind to antigens in the sample, forming antigen-antibody complexes, and migrate onto nitrocellulose matrix to be captured by the other immobilized-antibodies on test strip.

More antigens in the sample will form more antigen-antibody complexes which lead to stronger fluorescence signal by detector antibodies, which is processed by instrument for ichroma™ tests to show the content of glycated hemoglobin in terms of percent of the total hemoglobin in the blood.

COMPONENTS

ichroma™ HbA1c consists of 'cartridges', 'detection buffers', and 'a hemolysis buffer'.

- The cartridge contains a test strip, the membrane which has anti human HbA1c at the test line, and rabbit IgG at the control line. All cartridges are individually sealed in an aluminum foil pouch containing a desiccant.
- The detection buffer contains anti human hemoglobin A0-fluorescence conjugate, goat anti-rabbit IgG-fluorescence conjugate, bovine serum albumin (BSA) as a stabilizer and sodium azide in phosphate buffered saline (PBS) as a preservative.
- The hemolysis Buffer contains triton X-100 in DDW.
- The detection buffers are pre-dispensed in tubes. The detection buffers and hemolysis buffer are packaged in a box and further packed in a Styrofoam box with ice-pack for the shipment.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Follow instructions and procedures described in this 'Instruction for use'.
- Use only fresh samples and avoid direct sunlight.
- Lot numbers of all the test components (cartridge, detection buffer, hemolysis buffer and ID chip) must match each other.
- After using the hemolysis buffer, keep it closed.
- Do not interchange the test components between different lots or use the test components after the expiration date, either of which might yield incorrect of test result(s).
- Do not reuse cartridges or detection buffers. A cartridge should be used for testing one sample only. A detection buffer tube should be used for processing of one sample only.
- The cartridge should remain sealed in its original pouch until just before use. Do not use the cartridge, if pouch is damaged or has already been opened.
- Frozen sample should be thawed only once. For shipping, samples must be packed in accordance with the regulations. Sample with severe hemolysis and/or hyperlipidemia must not be used.
- Allow cartridge, detection buffer and sample to be at room temperature for approximately 30 minutes before use.
- The instrument for ichroma™ tests may generate slight vibration during use.
- Used cartridges, detection buffers and pipette tips should be handled carefully and discarded by an appropriate method in accordance with relevant local regulations.
- The mixture of detection buffer and hemolysis buffer must be used within 1 hour after mixing.
- An exposure to larger quantities of sodium azide may cause certain health issues like convulsions, low blood pressure and heart rate, loss of consciousness, lung injury and respiratory failure.
- **ichroma™ HbA1c** will provide accurate and reliable results subject to the below conditions.
  - **ichroma™ HbA1c** should be used only in conjunction with instrument for ichroma™ tests.
  - Have to use recommended anticoagulant sample.

Recommended anticoagulant
K_2EDTA, K_3EDTA, Na_2EDTA, Lithium heparin, Sodium citrate

STORAGE AND STABILITY

Component	Storage condition: Storage Temperature	Shelf life	Note
Cartridge	4-30 °C	20 months	Disposable
Detection buffer	2-8 °C	20 months	Disposable
Hemolysis buffer	4-30 °C	20 months	Unopened
	4-30 °C	12 months	Opened

- After the cartridge pouch is opened, the test should be performed immediately.

LIMITATIONS OF THE TEST SYSTEM

- The test may yield false positive result(s) due to the cross-reactions and/or non-specific adhesion of certain sample components to the capture/detector antibodies.
- The test may yield false negative result(s) due to the non-responsiveness of the antigen to the antibodies which is the most common if the epitope is masked by some unknown components, so therefore not being able to be detected or captured by the antibodies. The instability or degradation of the antigen with time and/or temperature may also cause false negative result as it makes antigen unrecognizable by the antibodies.
- Other factors may interfere with the test and cause erroneous results, such as technical/procedural errors, degradation of the test components/reagents or presence of interfering substances in the test samples.
- Any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician including clinical symptoms and other relevant test results.
- The test environment conditions for **ichroma™ HbA1c** are below.
  - Temperature: 20-30 °C
  - Humidity: 10-70 %
  - i-chamber target temperature: 30 °C

MATERIALS SUPPLIED

REF CFPC-38

Components of **ichroma™ HbA1c**

- Cartridge Box:
  - Cartridge 25
  - ID Chip 1
  - Instruction for Use 1
- Detection Buffer Box
  - Detection buffer tube 25
  - Hemolysis Buffer Vial (3 mL) 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ HbA1c**. Please contact our sales division for more information.

- Instrument for ichroma™ tests
  - ichroma™ Reader REF FR203
  - ichroma™ II REF FPRR021
- i-Chamber REF FPRR009
- Printer REF FPRR007
- Boditech HbA1c Control REF CFPO-96
- 5 µL Capillary tube REF CFPO-19

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ HbA1c** is human whole blood.

- It is recommended to test the sample within 12 hours after collection.
- Samples may be stored for a week at 2-8 °C prior to being tested. If testing will be delayed more than a week, samples should be frozen at -70 °C.
- Samples stored frozen at -70 °C for 3 months showed no performance difference.
- Once the sample was frozen, it should be used one time only for test, because repeated freezing and thawing can result in the change of test values.

TEST SETUP

- Check the components of the **ichroma™ HbA1c**: Cartridges, Detection buffers, a Hemolysis buffer, ID chip and Instruction for use.
- Ensure that the lot number of the test cartridges match that of detection buffers, hemolysis buffer as well as an ID chip.
- If the sealed cartridge and the detection buffer have been stored in a refrigerator, place them on a clean and flat surface at room temperature for at least 30 minutes before testing.
- Temperature of i-chamber should be 30 °C.

TEST PROCEDURE

1) Take out a cartridge from the pouch and insert it into the i-Chamber (30 °C).
2) Draw 100 µL of hemolysis buffer and transfer it into detection buffer tube.
3) Draw 5 µL of whole blood or control using 5 µL capillary tube and put the capillary tube into the detection buffer tube. (Do not make air bubbles in the capillary tube and careful not to get blood on the surface of the capillary tube. If blood gets on the surface of the capillary tube, remove it gently with gauze.)
4) Close the lid of the detection buffer tube and mix the sample thoroughly by shaking it about 15 times.
5) Take out the half of the cartridge from i-Chamber slot.
6) Pipette out 75 µL of the sample mixture and load it into a sample well of the test cartridge.
7) Wait till the sample mixture flow appears in the windows. (about 10 seconds)
8) Insert the cartridge into i-Chamber slot (30 °C).
9) Leave the cartridge in i-Chamber for 12 minutes before removing.
   ⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inaccurate test result.*
10) To scan the sample-loaded cartridge, insert it into the cartridge holder of the instrument for ichroma™ tests. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.
11) Press 'Select' button on the instrument for ichroma™ tests to start the scanning process.
12) Instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
13) Read the test result on the display screen of the instrument for ichroma™ tests.

INTERPRETATION OF TEST RESULT

- Instrument for ichroma™ tests calculates the test result automatically and displays HbA1c concentration of the test sample in terms of % (NGSP), mmol/mol (IFCC), mg/dL (eAG).
- The cut-off (reference range)
  - NGSP (%): 4.5-6.5 %
  - IFCC (mmol/mol): 26-48 mmol/mol
- Working range
  - NGSP (%): 4-15 %
  - IFCC (mmol/mol): 20.2-140.4 mmol/mol
  - eAG (mg/dL): 68.1-383.8 mg/dL

QUALITY CONTROL

- Quality control tests are a part of the good testing practice to confirm the expected results and validity of the assay and should be performed at regular intervals.
- The control tests should be performed immediately after opening a new test lot to ensure the test performance is not altered.
- Quality control tests should also be performed whenever there is any question concerning the validity of the test results.
- Control materials are provided on demand with **ichroma™ HbA1c**. For more information regarding obtaining the control materials, contact Boditech Med Inc.'s Sales Division for assistance. (Please refer to the instruction for use of control material.)

PERFORMANCE CHARACTERISTICS

- **Analytical Specificity**
  - Cross-reactivity

    Biomolecules such as below the ones in the table were added to the test sample(s) at concentrations much higher than their normal physiological levels in the blood. **ichroma™ HbA1c** test results did not show any significant cross-reactivity with these biomolecules.

Cross-reactivity material	Standard material conc. 5.2 %	6.5 %	10.5 %
	Recovery (%)		
HbA0 (20 mg/mL)	99.9	96.1	99.0
HbA1a,A1b (20 mg/mL)	100.9	96.8	101.0
Acetylated hemoglobin (100 mg/mL)	101.0	98.4	99.7
Carbamylated hemoglobin (100 mg/mL)	100.5	97.8	100.0
Glycated h-Albumin (100 mg/mL)	100.3	97.4	100.6
HbA1d (100 mg/mL)	100.9	97.0	100.3
Acetylaldehyde hemoglobin (100 mg/mL)	100.8	95.6	99.1

  - Interference

    Interference materials such as below the ones in the table were added to the test sample(s) the same as the below concentrations. **ichroma™ HbA1c** test results did not show any significant interference with these materials.

Interference material	Standard material conc. 5.6 %	6.5 %	10.5 %
	Recovery (%)		
Non-interference	101.0	96.2	98.7
Acetaminophen (20 mg/dL)	100.4	97.8	100.9
L-ascorbic acid (500 mg/dL)	101.0	97.8	99.8
Bilirubin (2 g/dL)	100.8	97.8	100.4
D-glucose (1,000 mg/dL)	100.9	97.6	99.8
Intralipid (8,000 U/L)	100.8	96.2	100.6
Triglyceride (327 M)	100.9	96.1	99.6
Urea (10 g/dL)	100.1	98.1	99.7

- **Precision**
  - Intra-assay

    The intra-assay precision was calculated by one evaluator who tested each concentration of different concentration of control standard five times each with three different lots of **ichroma™ HbA1c.**

HbA1c (%)	Lot 1	Lot 2	Lot 3	AVG	SD	CV (%)
5.2	5.28	5.18	5.24	5.23	0.12	2.36
6.5	6.46	6.48	6.34	6.43	0.13	1.99
10.5	10.4	10.56	10.58	10.51	0.19	1.83

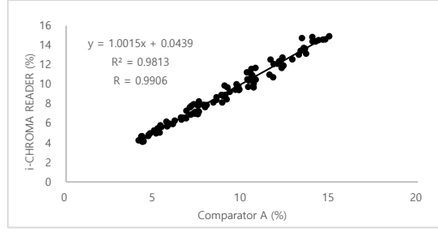
  - Inter-assay

    The inter-assay precision was confirmed by 3 different evaluators with 3 different lots, testing five times each different concentration.

HbA1c (%)	Between-person AVG	SD	CV (%)	Between-lot AVG	SD	CV (%)
5.2	5.19	0.03	0.61	5.23	0.05	0.96
6.5	6.51	0.02	0.36	6.43	0.07	1.12
10.5	10.50	0.01	0.10	10.51	0.10	0.92

- **Comparability:**

  HbA1c concentrations of 100 clinical samples were quantified independently with **ichroma™ HbA1c (i-CHROMA READER)** and Comparator A as per prescribed test procedures. Test results were compared, and their comparability was investigated with linear regression and coefficient of correlation (R). Linear regression and coefficient of correlation between the two tests were Y=1.0015x + 0.0439 and R = 0.9906 respectively.



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Note: Please refer to the table below to identify various symbols.

Σ	Sufficient for <n> tests
	Read instruction for use
	Use by Date
LOT	Batch code
REF	Catalog number
	Caution
	Manufacturer
EC REP	Authorized representative of the European Community
IVD	In vitro diagnostic medical device
	Temperature limit
	Do not reuse
CE	This product fulfills the requirements of the Directive 98/79/EC on in vitro diagnostic medical devices

CE IVD

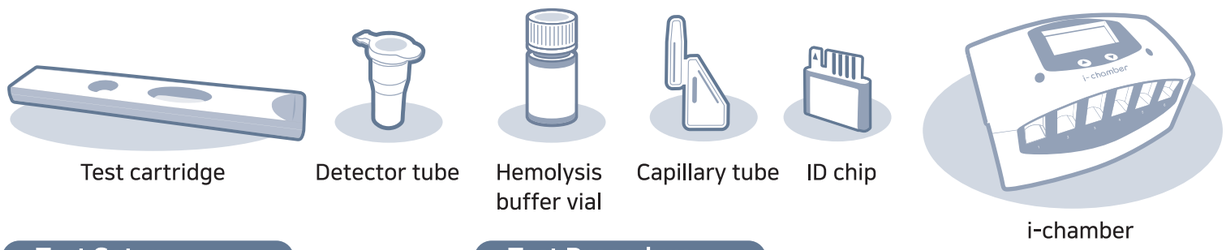
ichroma™

HbA1c

This is not a complete Instruction for use. For more detailed instructions, please refer to IFU.

HbA1c_Rev01_200706E_W

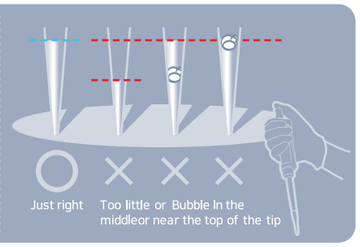
Test Components



Ensure that the lot number of the components match.

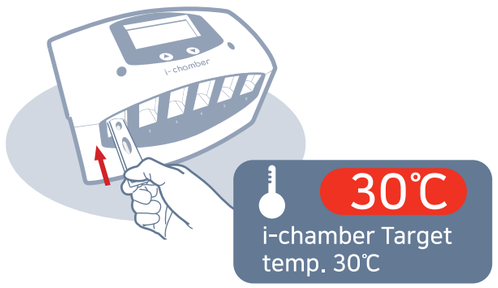
LOT Matches LOT

If extraction buffer tube are stored in refrigerator, keep the sample collection tube at room temperature for 30 minutes just prior to the test.



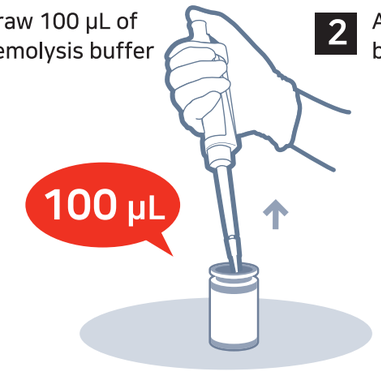
Test Setup

1 Insert a cartridge into an i-chamber slot

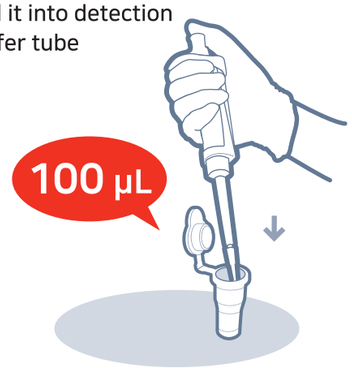


Test Procedure

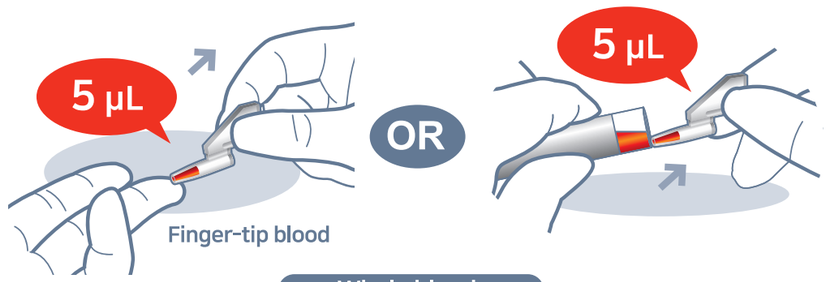
1 Draw 100 µL of hemolysis buffer

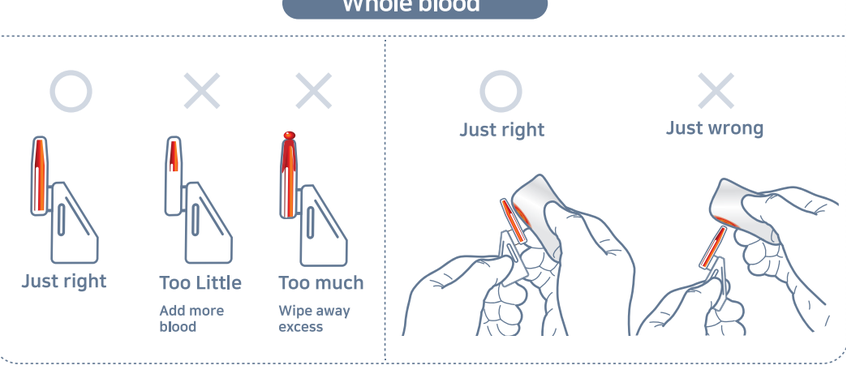


2 Add it into detection buffer tube

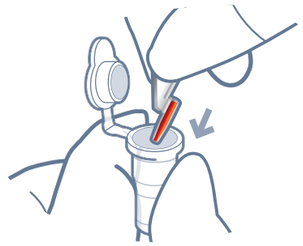


3 Draw whole blood



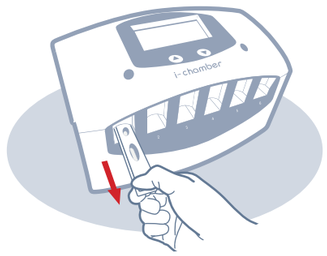


4 Put the capillary tube into the detection buffer tube

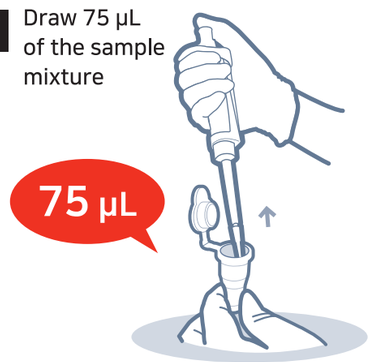


5 Shake the detection buffer tube

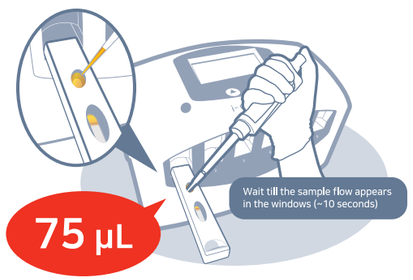
6 Take the cartridge half way out



7 Draw 75 µL of the sample mixture



8 Apply 75 µL in the sample well



9 Insert a cartridge into i-chamber and incubate for 12 minutes

10 Insert the test cartridge into i-CHROMA READER or ichroma™ II.

i-CHROMA READER

or

ichroma™ II

11 i-CHROMA READER : Press 'Select'
ichroma™ II : Tap 'Start'

i-CHROMA READER
Press **Select**

ichroma™ II
Tap **Start**

12 Read the test result

Result

For technical assistance; please contact: Boditech Med Inc. ' Technical Services '
Tel: +82-33-243-1400 Fax: +82-33-243-9373 E-mail: sales@boditech.co.kr
www.boditech.co.kr