HbA1c Rapid Test Kit
(Whole blood)

INTENDED USE:

The HbA1c Rapid Test Cassette (Whole Blood) is a rapid chromatographic Immunoassay for the semi-quantitative estimation of human HbA1c in whole blood is an aid in the monitoring of diabetes mellitus

For professional / self and in-vitro diagnostic use only.

ORDER INFORMATION

REF	Cont.
BBPHBC 01	01 Test
BBPHBC 05	05 Tests
BBPHBC 10	10 Tests
BBPHBC 25	25 Tests
BBPHBC 50	50 Tests

CLINICAL SIGNIFICANCE:

Glycated hemoglobin (HbA1c) is a form of hemoglobin that is measured primarily to identify the 3 months' average plasma glucose concentration. HbA1c is a measure of the beta-N-1 deoxy fructosyl component of hemoglobin. The test is limited to a 3 month average because the lifespan of a red blood cell is 4 months (120 days). However, since red blood cells do not all undergo lysis at the same time, HbA1c is taken as a limited measure of three months. It is formed in a non-enzymatic glycation pathway by hemoglobin's exposure to plasma glucose.

HbA1c is a measure of the beta-N-1-deoxy fructosyl component o hemoglobin. The origin of the naming derives from Hemoglobin type A being separated on cation exchange chromatography. The first fraction to separate, probably considered to be pure Hemoglobin A1 was Designated HbAo, the following fractions were designated HbA1c, HbA11b, HbA1c, respective of their order of elution. There have subsequently been many more sub fractions as separation techniques have improved. 2 Normal levels of glucose produce a normal amount of glycated hemoglobin. As the average amount of plasma glucose increases, the fraction of glycated hemoglobin increases in a predictable way. This serves as an indicator that blood sugar is increasing and that action should be taken.

In diabetes mellitus, higher amount of glycated hemoglobin, indicating poorer control of blood glucose levels, have been associated with cardiovascular disease, nephropathy, neuropathy, and retinopathy. A trial on a group of patients with Type 1 diabetes found that monitoring by caregivers of HbA1c led to changes in diabetes treatment and improvement of metabolic control compared to monitoring only of blood or urine glucose. However, a trail designed specifically to determine whether reducing HbA1c below the normal 6%, using primarily insulin and sulfonylureas (both know to easily drive blood sugar too low), and would reduce the rate of cardiovascular events in type 2 diabetes found higher mortality-trial was terminated early. The negative outcomes may well have been result of the treatment approach, primarily insulin and sulfonylureas, utilized in the "intensive" treatment group instead of LCHF (Low-Carbohydrate High Fat diet), GIP-1 analogues & SGLT-2 inhibitors. None of which have these problems & lower cardiovascular mortalities.

PRINCIPLE:

The HbA1c Rapid Test Cassette (Whole Blood) is a semi quantitative, membrane based Immunoassay for the detection of HbA1c in human whole blood. The membrane is pre-coated with anti-hemoglobin antibodies. During resting, the HbA1c in whole blood specimen reacts with anti-HbA1c part of the dye conjugate, which has been impregnated on the conjugate pad. The mixture then migrates upward on the membrane by capillary action, reacts with anit-hemoglobin on the membrane on Test Line region. If the specimen contains HbA1c, a colored line will appear in test line region. The absence of the colored lines in test line region indicates that the specimen does not contain HbA1c, or the concentration of HbA1c is lower than cut-off value. To serve as a procedure control, a colored line will always appear in the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

COMPONENTS:

1. Individually foiled Test device with desiccant
2. Prefill Assay buffer
3. Sample Dispense dropper (25 µl sampling device)
4. Instruction for use
5. Sample collection Dropper 5µl (sample addition)
6. Blood lancet
7. Alcohol swab
8. Color chart

STORAGE AND STABILITY:

1. The kit can be stored at room temperature or refrigerated (2-30°C). The test device must remain in the sealed aluminum pouch until use. DO NOT FREEZE.
2. Do not use beyond the expiration date.
3. Do not use the test device/strip, if the pouch is damaged or seal is broken.

PRECAUTIONS

1. Wear protective gloves while handling specimens wash thoroughly afterwards.
2. The device is sensitive to humidity as well as heat. Therefore, take out the device from seal pouch before test.
3. Do not mix reagents from different lot.
4. Dispose all the samples and kits properly as per the instruction after test in accordance in GLP.
5. Follow the testing procedure exactly as mention in the insert.

LIMITATIONS:

1. The HbA1c Rapid Test Kit is for in vitro diagnostic use only. Results should be interpreted in conjunction with other clinical and laboratory findings.
2. The test may provide false-positive or false-negative results. Confirmatory testing should be conducted if necessary.
3. Interference from substances such as rheumatoid factor, heterophilic antibodies, or lipemic or hemolyzed specimens may affect the test performance.
4. This test is optimized for human blood samples. Other sample types or interference may affect the results.

SPECIMEN COLLECTION AND PREPARATION:

1. Collect a fresh whole blood sample using standard laboratory procedures.
2. Remove any particulate matter or precipitate by centrifugation before testing if required.
3. Avoid hemolysis, as it may interfere with the test results.
4. **Whole Blood (WB):** Use Whole Blood samples collected by venipuncture into a collection tube containing EDTA, citrate or heparin and should be store 2-8°C if the test is to be use within 3 day of collection. Do not Freeze whole blood specimens. Alternately, collect the whole blood by lancing devices. WB can be delivered by pipette or sample dropper directly to the test card.

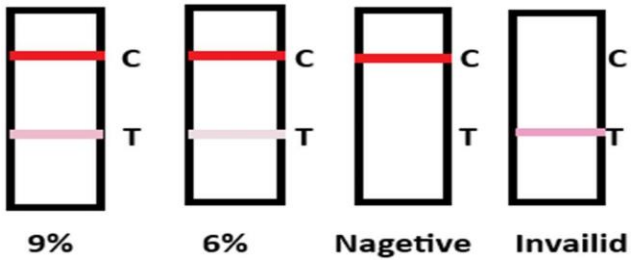
TEST PROCEDURE:

1. Bring the test device, sample buffer, and specimens to room temperature (15-30°C) before use.
2. Open the pouch and place the test device on a clean and flat surface.

3. Add 5µl whole blood sample in prefill assay buffer after that mix 10 times and incubate 2 mins.
4. Dispense 3 drops (approximately 75µL) of the diluted sample into the sample well of the test device using the sample dropper provided with the kit.
5. Start the timer and wait for the colored bands to appear.
6. Read the test results within 15 minutes. Do not interpret the results after 20 minutes.

INTERPRETATION OF RESULTS:

1- **Positive:** Two distinct red bands appear, one in the test region (T) and the second one in the control region (C).
Note: The intensity of the color in the test line region (T) will vary depending on the concentration of the HbA1c present in the specimen. **Read the positive result with the help of color chart provided in the kit as your reference to confirm the concentration of HbA1c in the whole blood sample.**
2- **Negative:** Only one red band appears in the control region (C). No band appears in the test region (T) or the test line is weaker than the reference line representing 6 % on the color chart. This indicates HbA1c to be healthy level meaning excellent control.
3- **Invalid:** If no red band appears in the control region (C), the test is invalid. Repeat the test with a new device.



Quality Control

Internal procedural controls are included in the test individually. A colored line appearing in control line region (C) is the internal procedural control. It confirms sufficient specimen volume and correct procedural technique. Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

PERFORMANCE CHARACTERISTICS

Detection Limitation
The HbA1c Rapid Test Cassette (Whole Blood) can detect HbA1c as low as 6 %.
Sensitivity and Specificity
The HbA1c Rapid Test Cassette (Whole Blood) was compared with commercial HbA1c ECLIA kit; the results indicate that HbA1c Rapid Test Cassette (Whole Blood) has a high sensitivity and specificity.

Method		ECLIA		Total results
HbA1c Rapid Test Cassette (Whole Blood)	Results	Positive	Negative	
	Positive	69	1	70
	Negative	1	99	100
Total Results		70	100	170

Relative Sensitivity: > 98.6% (95%CI*:96.1%-100%)
*Confidence Interval
Relative Sensitivity: 98.7% (95%CI*: 95.3%-99.8%)
Overall Accuracy: 99.1% (95%CI*: 96.8%-99.9%)

Precision

Intra-Assay

Within-run precision has been determined by using 3 replicates of e following specimens: negative, 6% Hba1c ad 9% Hba1c. The negative, 6% HbA1c and 9%HbA1c values were correctly identified >99% of the time.

Inter-Assay

Between-run precision has been determined by 3 independent assays on the same specimens: negative. 6% HbA1c and 9% HbA1c positive specimen. Three different lots of the HbA1c Rapid Test Cassette (Whole Blood) have been tested over a 3-days period using negative, 6% HbA1c and 9% HbA1c positive specimens. The specimens were correctly identified >99% of the time.

Cross-reactivity

The HbA1c Rapid Test Cassette (Whole Blood) has been tested for HbsAg, anti-HIV, anti-HCV, anti-RF, anti-Spyhilis, anti-H.pylori, anti-Toxo IgG, anti-CMV IgG positive specimens. The results showed no cross-reactivity.

INTERFERING SUBSTANCES

The Following compounds have also been tested using the HbA1c Rapid Test Cassette (Whole Blood) and no interference was observed.
Acetaminophen: 20mg/dl caffeine: 20mg/dl
Creatinine: 200mg/dl
Acetylsalicylic Acid: 20mg/dl Gentisic Acid: 20mg/dl Albumin: 2000mg/dl
Ascorbic Acid: 2g/dl Hemoglobin: 1000mg/dl
Oxalic Acid: 600mg/dl
Bilirubin: 1000mg/dL

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3. Larsen ML, Horder M, mogensen EF (1990). "Effect of long-term monitoring of glycosylated haemoglobin levels In insulin-dependent diabetes mellitus". N. Engl. J. Med. 323 (15): 1021-25.
4. Gerstein HC, Mileer ME, Byington RP, et al. (2008). "Effects of intensive Glucose Lowering in Type 2 Diabetes". New England Journal of Medicine. 358(24): 2545-59

GLOSSARY OF SYMBOL

	Consult Instruction for Use
REF	Catalog Number
	Store between
	Manufacturer
	Keep away from sunlight

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