TECO DIAGNOSTICS
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HbA1c DIRECT REAGENT SET
TC MATRIX-240/480

INTENDED USE
The Teco HbA1c assay is used in clinical laboratories for the quantitative in vitro measurement of HbA1c (hemoglobin fraction) in human whole blood on auto analyzers.

INTRODUCTION[1,2,3]
The HbA1c assay is used as an aid in the monitoring of long-term blood glucose control and compliance in individuals with diabetes mellitus.

The Teco HbA1c assay is used for the measurement of the concentration of HbA1c relative to the concentration of the total hemoglobin (THb).

An increase in the percentage of HbA1c has been found in individuals diagnosed with diabetes. Uncontrolled diabetes can lead to acute complications such as cardiovascular disease, retinopathy, nephropathy, and neuropathy can occur. According to several studies, including the findings of Diabetes Control and Complications Trial (DCCT), these complications can be prevented by long-term control of diabetes.

Therefore, measurement of HbA1c percentage can be invaluable in the monitoring of long-term glycemic control of diabetic patients.

Teco HbA1c assay has no cross reactivity with labile HbA1c since the antibody used in this assay is specific for the ketoamine form of HbA1c. Stable HbA1c does not increase or decrease in response to rapid changes in physiological factors and therefore allows the measurement of individuals' average blood glucose levels over several months.

It is a liquid monoreagent. Store at 5 - 8°C. For in Vitro Diagnostic Use Only (IVD). Do not freeze.

Correlation between HbA1c and daytime glucose levels		
HbA1c (%)	Gluc. (mg/dL)	Gluc. (mmol/L)
5	97	5.4
6	126	7.0
7	154	8.6
8	183	10.2
9	212	11.8
10	240	13.4
11	269	14.9
12	298	16.5
Values are sourced from National Glycohemoglobin Standardization Program (NGSP) global website 2013-2014		

PRINCIPLE[4, 5]
Hemoglobin A1c is an important test recommended by the American Diabetes Association (ADA) and its usefulness was clarified by the United Kingdom Prospective Diabetes Study (UKPDS) and Diabetes Control and Complications Trial (DCCT). Currently, the HbA1c test is recommended for patients diabetes every 2-3 months as part of the patient diabetes management program. Glycohemoglobin is produced by non-enzymatic addition of glucose to amino groups in hemoglobin. HbA1c refers to glucose modified hemoglobin A (HbA) specifically at N-terminal valine residues of hemoglobin beta chains. HbA1c test is used both as an index of mean glycemia and as a measure of risk for the development of diabetes complications. Therefore, the HbA1c test is a good indicator of glycemic control in the preceding 2-3 months.

This method utilizes the interaction of antigen and antibody to directly determine the HbA1c in whole blood. Total hemoglobin and HbA1c have the same unspecific absorption rate to latex particles. When mouse antihuman HbA1c monoclonal antibody is added (R2), latex-HbA1c-mouse anti human HbA1c antibody complex is formed. Agglutination is formed when goat antimouse IgG polyclonal antibody interacts with the monoclonal antibody. The amount of agglutination is proportional to the amount of HbA1c absorbed on to the surface of latex particles. The amount of agglutination is measured as absorbance.

REAGENT COMPOSITION
1. Lyse Reagent: Stabilizers buffers, Lysing agent and water
2. Reagent R1: Latex: < 0, 15% Buffer Stabilizers
3. Reagent R2: Mouse anti-human HbA1c monoclonal antibody < 0.06 mg/mL, goat anti-mouse IgG polyclonal antibody < 0.09 mg/dL, Buffer and Stabilizers.

REAGENT DETERIORATION
If turbidity has occurred, turbidity may be a sign of contamination.

WARNING AND PRECAUTIONS
1. The reagent is for *in vitro* diagnostic use. Do not use expired reagents. Reagents with two different lot numbers should not be interchanged.
2. For professional use.
3. Follow Good Laboratory Practice (GLP) guidelines.
4. Contains sodium azide.

CAUTION: Human source samples are processed with this product. All human source samples must be treated as potentially infectious materials and must be handled in accordance with OSHA standards.

STORAGE AND STABILITY[6]
Store reagent set at 2 - 8°C till the expiration date stated on the label which is only for closed vials. Once opened vials are stable for 30 days at 2 - 8°C in optimum conditions. On board stability is strongly related to auto analyzers' cooling specification and carry-over values.

Reagent stability and storage data have been verified by using Clinical and Laboratory Standards Institute (CLSI) EP25-A protocol.

SPECIMEN COLLECTION
The assay is formulated for use with human whole blood samples. Venous whole blood samples collected with EDTA anticoagulant can be used. It is recommended that samples be used within 7 days of collection when stored refrigerated. Prior to testing, whole blood

samples should be mixed by gentle inversion to re-suspend settled erythrocytes.

Auto analyzer usage: Samples should be tested by stat mode (Emergency mode) to avoid precipitation.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Test tubes/rack.
2. Paper towel
3. gloves

PROCEDURES

Preparation of Hemolysate

1. Whole blood samples are taken to room temperature.
2. Blood samples are mixed in order to mix erythrocytes homogeneously.
3. Using a calibrated pipette, transfer 1000mL Lyse solution to the sample cup.
4. 30 mL of homogenized blood sample is transferred to the sample cup with Lyse added.
5. Hemolysate is mixed thoroughly, incubated for 5 mins at room temperature.
6. Hemolysate is ready for use for HbA1c.

Test Name:	HbA1c	R1:	118
Full Name:	HbA1c	R2:	34
Pri. Wave:	670 nm	SAMPLE VOLUME:	3.5
Sec. Wave:	/	Calibration Type:	Spline
Assay/ Point:	Fixed Time	K Value:	/
Start - End:	18 - 30	Point:	/
Decimal place:	1	Blank Type:	Reagent
Unit:	%	Point 0 (Blank) Con.:	0.0
Linearity Range:	/ - /	Point 1 (Cal) Con.: calibrator/ standard	
		Point 2 (Cal) Con.: calibrator/ standard	
		Point 3 (Cal) Con.: calibrator/ standard	
Correlation Factor: 1.0000 - 0.0000		Point 4 (Cal) Con.: calibrator/ standard	

QUALITY CONTROL

Teco Turbidimetric HbA1c assay requires monthly calibration. Place calibration series on the analyzer in the order of lowest to highest.Enter calibrator lot specific values provided on the specification sheet.

The assay requires the use of an HbA1c calibrator 4-levels. We recommend HbA1c Calibrator 4-Level.

EXPECTED VALUES

4.5-6.5% (NGSP/DCCT)
26-48 mmol/mol (IFCC)
Levels above 6.5% HbA1c are suitable for the diagnosis of diabetes mellitus according to the data provided by NGSP. Patients with levels between 39-46 mmol/mol (IFCC) or 5.7-6.4% HbA1c (NGSP) have a possibility of developing diabetes risk.[9,10]

It is recommended that each laboratory establish its own normal range.

Reference interval has been verified by using CLSI EP28- A3c protocol.

CALIBRATION STABILITY

All calibrator vials are stable until their expiration date when stored at 2 - 8°C.

Commercially available control material with established values determined by this method may be used. We recommend HbA1c Control- Low/ High Set

PERFORMANCE CHARACTERISTICS

1. Linearity: 15%
2. Limit of Quantitation (LoQ): LoQ values are based on Coefficient of Variation Percentage (CV) 20%[8]: 4%
3. Precision Study[12]:

	Within Run	
Mean (%)	C.V.%	n
5.46	1.45	40
10.1	1.73	40

	Run to Run	
Mean (%)	C.V.%	n
5.46	2.81	40
10.1	2.71	80

Precision Studies data have been verified by using CLSI EP05-A3 protocol.

4. Interference[3, 4, 15]: No significant interactions were observed for conjugated bilirubin, triglycerides, ascorbic acid, acetlated Hb, carbamylated Hb up to the interferent concentration given in the table.

Interferant	Concentration
Ascorbic acid	40 mg/dL
Total bilirubin	48 mg/dL
Acetylated Hb	4.8 mmol/L
Triglycerides	2000 mg/dL
Carbamylated Hb	7.3 mmol/L

Stable glycated hemoglobin serves as a substrate for the Turbidimetric reaction used in Teco Direct Turbidimetric HbA1c assay.

The acceptable interference limit is set 10% below the highest interference concentration within ±10% recovery of the target.

Interferences may affect the results due to medication or endogenous substances.

These performance characteristics have been obtained by using an analyzer. Results may vary if a different instrument or a manual procedure is used.

WARNINGS AND PRECAUTIONS

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