510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k102510

B. Purpose for Submission:

New device

C. Measurand:

Whole Blood Glycated Hemoglobin (HbA1c)

D. Type of Test:

Quantitative, turbidometric, inhibition immunoassay

E. Applicant:

Siemens Healthcare Diagnostics Inc.

F. Proprietary and Established Names:

Dimension® HB1C kit (Model DF 105A)

Dimension® HB1C Calibrator

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
LCP	Class II	21 CFR 864.7470	Hematology (81)
JJX	Class II	21 CFR 862.1150	Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See indications for use below.

2. Indication(s) for use:

   The Dimension® HB1C assay is an *in vitro* diagnostic assay for the quantitative determination of hemoglobin A1c (Hb A1c) in human anticoagulated whole blood on the Siemens Dimension RxL instrument. Measurements of HbA1c are used for monitoring long-term glycemic control in individuals with diabetes mellitus.

   The HB1C CAL is an *in vitro* diagnostic product for the calibration of the Hemoglobin A1c (HB1C) method on the Dimension® clinical chemistry system.

3. Special conditions for use statement(s):

   For prescription use only

   The sponsor has the following limitation statements in the labeling:

   "Any cause of shortened red cell survival will reduce the exposure of red cells to glucose with a consequent decrease in HB1C values, e.g. hemolytic anemia or other hemolytic diseases, pregnancy, recent significant blood loss, etc. Results of HbA1c are not reliable in patients with chronic blood loss and consequent variable erythrocyte lifespan."

   "The antibody reagent used in the HB1C method will measure any glycosylated hemoglobin variants that are glycated at the beta-chain N-terminus and have epitopes identical to that of HbA1c (amino acid sequence: VAL-HIS-LEU-THR). This includes HbS, HbG, HbH, Hb Wayne, HbC, HbE, etc. Other hemoglobinopathies may give incorrect results with this test. Care must be taken when interpreting any HbA1c result from patients with Hb variants. Abnormal hemoglobins might affect the half life of the red cells or the in vivo glycation rates. In these cases, even analytically correct results do not reflect the same level of glycemic control that would be expected in patients with normal hemoglobin."

   "Glycated HbF (Fetal Hemoglobin), consisting of two alpha and two gamma chains, is not recognized by the anti-HbA1c antibody. Individuals with elevated levels of HbF (> 10%), most commonly found in infants and in patients with beta thalassemia, may produce lower than expected results with this assay. These samples must be assayed by an alternate method."

4. Special instrument requirements:

   Dimension RxL Instrument

I. Device Description:

This *in vitro* diagnostic device contains reagents and calibrator materials as follows:

Reagents:

Kit Well	Ingredient	Concentration
1, 2	Antibody (Sheep, polyclonal from ovine serum)	≥ 0.5 mg/mL
	2-morpholinoethane sulfonic acid (MES buffer)	0.025 M
	Tris(hydroxymethyl)-aminomethane (TRIS) buffer, pH 6.2	0.015 M
	Stabilizers	
3	Polyhapten Reagent	≥ 8 µg.mL
	MES buffer	0.25 M
	TRIS buffer, pH 6.2	0.015 M
	Stabilizers	
4	Empty	
5,6	tetradecyl trimethyl ammonium bromide (hemolyzing reagent)	< 1%

Calibrator materials:

The Dimension® HB1C kit contains 5 levels of calibrators that are lyophilized whole blood lysate containing various concentrations of HbA1c and total hemoglobin. There are 3 levels of calibration for total hemoglobin (levels 3-5) and 5 levels of calibration for HbA1c. To use, 2.0 mL Clinical Laboratory Reagent Water must be added to each vial, followed by gentle swirling.

The human blood used in the manufacture of these calibrators has been tested using FDA approved methods and found to be non-reactive for HBsAg and antibodies to HCV and HIV 1/2.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   BIO-RAD Variant® II HbA1c

2. Predicate 510(k) number(s):

   k070452

3. Comparison with predicate:

Similarities		
Item	Predicate	Candidate Device
Intended Use Kit	Same	For *in vitro* diagnostic use for the quantitative determination of hemoglobin A1c in human whole blood

Similarities		
Item	Predicate	Candidate Device
Intended Use Calibrator Material	Same	For *in vitro* diagnostic calibration of the Hemoglobin A1c method
Sample Type	Same	Human anticoagulated whole blood treated with EDTA
Calibrator Materials	Same	Lyophilized whole blood hemolysate
Certification	Same	NGSP certified as traceable to the DCCT
Traceability	Same	IFCC reference method
Sample Preparation	Same	Sample directly and dilute on board
Packaging	Same	Kit contains reagents and calibrators

Differences		
Item	Predicate	Candidate Device
Instrument	BIO-RAD Variant® II HPLC	Dimension® RxL clinical chemistry system
Reporting Units	% HbA1c	% HbA1c and mmol/mol Hb
Analytical Measuring Range	3.1 – 18.5 % HbA1c	3.6 – 16.0 % HbA1c (17-151 mmol/mol Hb)
Calibrator Levels	2 cal levels plus diluent for additional levels	5 cal levels
Technology	Ion-exchange high performance liquid chromatography	Turbidometric inhibition immunoassay for HbA1c and a modified alkaline hematin reaction for total Hb

K. Standard/Guidance Document Referenced (if applicable):

None were referenced

L. Test Principle:

The Dimension® HB1C assay measures both the HbA1c and the total hemoglobin in each sample, and reports the HbA1c either as % total hemoglobin or as mmol/mol total hemoglobin. Both measurements are automated.

Hemoglobin A1c Measurement: whole blood is treated with lysing reagent and is then mixed with the anti-HbA1c antibody in a buffered reagent. The HbA1c in the sample forms a soluble complex with the anti-HbA1c. A polyhapten reagent containing multiple HbA1c epitopes is then added to the sample, forming an insoluble complex with the excess free anti-HbA1c antibody. This antibody-polyhapten complex is then measured turbidimetrically at 340 nm.

Total Hemoglobin Measurement: whole blood is treated with lysing reagent which releases and converts the hemoglobin in the cell to a derivative that can be spectrophotometrically measured at 405 and 700 nm.

Two ratio calculation options are provided to the customer in the package insert. The first option reports the results in % HbA1c while the second option reports the results in SI units of mmol/mol total hemoglobin. There are several sub-options the customer can choose that calculates the % HbA1c in NGSP standardized results or can convert the mmol/mol results to % HbA1c using the IFCC-NGSP master equation.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

A precision study was performed by testing 4 levels of purchased whole blood hemolysate on the Dimension RxL instrument, assayed in duplicate with 2 runs per day for 20 days (n=80). Results were calculated for both % HbA1c and mmol/mol units.

Results Calculated from % HbA1c:

Sample	Mean (% HbA1c)	Within Run		Total Imprecision	
		SD	%CV	SD	%CV
Level 1	5.37	0.10	1.94	0.11	2.04
Level 2	6.12	0.07	1.13	0.10	1.58
Level 3	8.20	0.13	1.57	0.13	1.58
Level 4	11.87	0.15	1.30	0.17	1.40

Results Calculated from mmol/mol total hemoglobin:

Sample	Mean (mmol/mol HbA1c)	Within Run		Total Imprecision	
		SD	%CV	SD	%CV
Level 1	35.3	0.97	2.8	1.02	2.9
Level 2	43.2	0.71	1.6	1.02	2.4
Level 3	65.9	1.49	2.3	1.51	2.3
Level 4	107.0	1.77	1.7	2.01	1.9

A second precision study was performed using four whole blood pools with %HbA1c values across the assay range. Each pool was assayed in duplicate for 20 days and 2 runs per day, with 2 reagent lots, 2 instruments and a single calibration for each instrument (n=80). Results were calculated for both %HbA1c and mmol/mol units.

Results Calculated from % HbA1c:

Sample	Mean	Within Run	Total Imprecision

	(% HbA1c)	SD	%CV	SD	%CV
Pool 1	5.1	0.09	1.8	0.1	2.1
Pool 2	6.4	0.07	1.1	0.1	1.3
Pool 3	7.6	0.11	1.4	0.1	1.5
Pool 4	9.3	0.13	1.4	0.2	1.7

Results Calculated from mmol/mol total hemoglobin:

Sample	Mean (mmol/mol HbA1c)	Within Run		Total Imprecision	
		SD	%CV	SD	%CV
Pool 1	32	0.79	2.5	0.91	2.8
Pool 2	44	0.59	1.4	0.63	1.4
Pool 3	54	0.94	1.7	1.01	1.9
Pool 4	69	1.11	1.6	1.45	2.1

b. *Linearity/assay reportable range:*

Linearity was evaluated following CLSI guideline EP6-A. A high HbA1c sample of human whole blood (18.6% HbA1c /186 mmol/mol) and low HbA1c sample of whole blood material from a commercial source that contains a typical hemoglobin concentration and very low HbA1c concentration were used for this study. Dilutions were prepared from these two samples to provide various concentrations of HbA1c while maintaining a relatively constant hemoglobin level. The HbA1c concentrations ranged from 3.6% HbA1c to 16% HbA1c.

Statistical evaluations using linear regression showed that the assay is linear from 3.6% HbA1c to 16% HbA1c, yielding a linear regression analysis of $y = 0.97x - 0.19$, $r = 1.00$.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

Traceability

The assigned HbA1c and total hemoglobin values of the calibrators are traceable to the NGSP reference method through a correlation study. The Dimension Hemoglobin A1c assay is certified with the National Glycohemoglobin Standardization Program (NGSP). The NGSP certification expires in one year. See NGSP website for current certification at http://www.ngsp.org.

The derived result of the ratio (%) from the NGSP correlation is calculated from the individual quantitative results for total hemoglobin and Hemoglobin A1c (HbA1c). The International Federation of Clinical Chemistry (IFCC) units of mmol/mol are calculated using the Master Equation:

IFCC = (NGSP- 2.15) / 0.092

Two different units are provided to the customers:

NGSP equivalent units (%) and IFCC equivalents units (mmol/mol)

Value Assignment

The initial lot of calibrator materials (master lot) is value assigned through a correlation study using samples provided by the NGSP with values determined on the NGSP reference method. Based on the correlation study with the NGSP Reference method, the total hemoglobin and HbA1c values for the Master Lot are established. All subsequent lots are value assigned against the Master Lot.

Stability

The shelf life, freeze-thaw, stress, and open vial stability testing protocols for the calibrator materials and the acceptance criteria were described and found to be acceptable. Calibrator materials are stable until expiration date (12 months) when stored at 2-8ºC. After reconstitution, calibrators are stable for 8 hours at room temperature or for 48 hours at 2-8ºC.

Reagent stability protocol and acceptance criteria was also evaluated and found to be acceptable. Reagents are stable in storage at 2-8ºC for 12 months.

Sample stability was investigated and the protocol and acceptance criteria were found to be acceptable. Specimens are stated to be stable when stored for no greater than 3 days at 15 – 25 °C, 7 days at 2 – 8 °C, and 4 months at -20 °C (freeze only once).

A calibration interval study protocol and acceptance criteria was described and found to be acceptable. The stated calibration interval is 30 days.

d. Detection limit:

The limit of the blank (LoB) for both Hb and HbA1c was determined from 60 measurements of 5 blank samples (EDTA plasma from 3 healthy individuals mixed 1:1 with a commercially available whole blood material that contains normal Hb concentration and very low HbA1c concentration) on 2 separate Dimension RxL instruments. The LoB was calculated to be 2.6 % HbA1c (7.0 mmol/mol HbA1c).

The limit of detection (LoD) was determined from 96 measurements from 8 low samples (EDTA whole blood diluted with EDTA plasma) on 2 separate

Dimension RxL instruments. The LoD was calculated to be 3.6 % Hb A1c (17 mmol/mol Hb A1c).

e. Analytical specificity:

To test for interference by hemoglobin variants, samples were screened and identified as containing a % of each hemoglobin variant of interest by the Diabetes Diagnostic Laboratory (DDL) at the University of Missouri. The HbA1c value was determined by an NGSP recognized reference method. Two different sets of samples were prepared for each variant study by mixing a high variant sample with a low/no Hb variant sample. Eight or nine of these dilution samples were prepared, bringing the total number of samples for the study to ten or eleven. An expected HbA1c value was assigned to each sample based on the values determined by an NGSP recognized reference method for variants HbC, HbD, HbE and HbS, and using the ion-exchange HPLC for Hb F, and the mixing ratio of high and low samples. The Dimension® HB1C method was used to assay each sample (n=5). The mean observed value was compared with the expected % HbA1c for each sample, and no interference was defined as less than 10% difference in recovery versus the low/no variant sample. Results for the interference testing of HbC, HbD, HbE and HbS showed no interference with the proposed assay, however HbF was shown to have significant interference above 10% HbF.

Studies were performed to assess common endogenous and exogenous substances that could interfere with the assay. Fifty-five potential interferents, including bilirubin, cholesterol, creatinine, ibuprofen, triglycerides, and urea, were tested by spiking into EDTA treated whole blood samples at two HbA1c concentrations. All percent biases as compared to the control (unspiked sample) were < 10%, and were deemed to have no significant interference with the assay results.

To test the level of interference of carbamylated, acetylated and labile hemoglobin, two aliquots from two patient EDTA whole blood specimen pools, one with a normal HbA1c concentration and one with a high HbA1c concentration, were treated with urea, acetaldehyde and glucose respectively. Treated sample aliquots were compared to untreated sample aliquots after triplicate measurements. All results met the stated acceptance criteria for significant interference of less than 10% bias between carbamylated, acetylated or labile hemoglobin samples and their respective control samples.

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

Two method comparison studies were performed. The first was a comparison to the predicate device, Bio-Rad Variant™ II Hemoglobin A1c, and was performed at a laboratory site. Briefly, 126 fresh, excess, de-identified EDTA-treated whole blood samples with HbA1c within the range of 4.4 - 15.8 % HbA1c were run in duplicate (data analyzed in singlicate) on the proposed device and compared to single measurements run on the predicate device. Testing was performed by a single MT(ASCP) operator with previous training on the operation of both analyzers.

A second method comparison study was performed versus the Dimension® HA1C method previously cleared under k011852 which has the same intended use and measuring principle. The purpose of this second method comparison was to show good comparison between the proposed new HbA1c assay and the previously marketed HA1C assay. The comparison study was performed in-house by technicians who were trained on the operation of the instruments, using 140 excess de-identified EDTA- treated whole blood samples purchased from a vendor. Each of the 140 de-identified whole blood samples which ranged from 5.0 - 14.8 % HbA1c, was assayed in duplicate by each method, however only the first replicate was included for linear regression analysis.

The results of linear regression analysis are summarized in the table below.

Study	N	Range (%HbA1c)	Slope	Intercept	r
Dimension® HB1C kit vs. Predicate	124	4.4-16.2	0.90	0.44	0.987
Dimension® HB1C kit vs. k011852	140	5.0-14.8	1.04	0.17	0.985

b. Matrix comparison:

Not applicable

3. Clinical studies:

a. Clinical Sensitivity:

Not applicable

b. Clinical specificity:

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable

4. Clinical cut-off**:**

   Not applicable

5. Expected values/Reference range:

   The listed expected normal values for the assay of 4.5 – 6.2 % Hb A1c (24 – 43 mmol/mol) were determined in whole blood specimens from a population of 183 (89 females, 94 males) healthy adults (age 19-84 years) who were included based upon a normal healthy blood glucose result (exclusion criteria: random glucose of > 200 mg/dL or a fasting blood glucose of > 126 mg/dL). The labeling states that each laboratory should check the validity of this reference range and if necessary establish its own patient population specific reference interval.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.