

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

**A. 510(k) Number:**

k070819

**B. Purpose for Submission:**

Modification to device

**C. Measurand:**

Hemoglobin A1c

**D. Type of Test:**

Quantitative High Performance Liquid Chromatography

**E. Applicant:**

Bio-Rad Laboratories, Inc

**F. Proprietary and Established Names:**

VARIANT II TURBO Link Hemoglobin Testing System

**G. Regulatory Information:**

1. Regulation section:

21CFR Section 864.7470 – Glycosylated Hemoglobin Assay

21CFR Section 864.8165 Calibrator for hemoglobin or hematocrit measurement.

2. Classification:

Class II

3. Product code:

LCP, KRZ

4. Panel:

Hematology (81)

**H. Intended Use:**

1. Intended use(s):

See indications for use below.

2. Indication(s) for use:

The Bio-Rad VARIANT™ II TURBO Link Hemoglobin A1c Program is intended for the percent determination of hemoglobin A1c in human whole blood using ion-exchange high-performance liquid chromatography (HPLC).

The VARIANT™ II TURBO Link Hemoglobin A1c Program is for use with the VARIANT™ II TURBO Link Hemoglobin Testing System interfaced with an automated sample transport system.

The Bio-Rad VARIANT™ II TURBO Link Hemoglobin A1c Program is for Professional Use Only.

Measurement of percent hemoglobin A1c is effective in monitoring long-term glucose control in individuals with diabetes mellitus.

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

VARIANT II TURBO Link Hemoglobin Testing System with an automated transport system [Sysmex® HST-N (Hemoglobin Transportation System) (k920544)].

**I. Device Description:**

The Bio-Rad VARIANT II TURBO Link Hemoglobin Testing System integrates the VARIANT II TURBO with an automated transportation system [Sysmex HST-N (Hematology Sample Transportation) System]. The VARIANT II TURBO Link Hemoglobin Testing System using Clinical Data Management (CDM) 4.1 software communicates with the Sysmex HST-N automation hardware and software in order to receive, identify, inject, and analyze samples.

The VARIANT II TURBO Link Hemoglobin Testing System is a fully automated,

high-throughput hemoglobin analyzer. It consists of three modules: The VARIANT II TURBO Link Chromatographic Station (VCS), the VARIANT II TURBO Link Sampling Station (VSS), and the Reagent Reservoir Module. In addition, a personal computer is used to control the VARIANT II TURBO Link Hemoglobin Testing System using Clinical Data Management (CDM) software version 4.1.

The VARIANT II TURBO Link Hemoglobin A1c Program contains Analytical cartridges, Guard cartridges, Elution Buffers A and B, Calibrator 1 and 2, Calibrator Diluent, Whole Blood Primer, Wash Buffer/Diluent Solution, a CD-ROM with program parameters to run 1600 tests.

**J. Substantial Equivalence Information:**

1. Predicate device name(s):

VARIANT II TURBO Hemoglobin A1c Program

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

k040872

3. Comparison with predicate:

<b>Similarities</b>		
<b>Item</b>	<b>Device</b>	<b>Predicate</b>
Assay Principle	Cation exchange high performance liquid chromatography	Cation exchange high performance liquid chromatography
Sample Type	Human anticoagulated whole blood (EDTA)	Human anticoagulated whole blood (EDTA)
Standardization	Traceable to the Diabetes Control and Complications Trial (DCCT) reference method and IFCC. Certified via the National Glycohemoglobin Standardization Program (NGSP).	Traceable to the Diabetes Control and Complications Trial (DCCT) reference method and IFCC. Certified via the National Glycohemoglobin Standardization Program (NGSP).

<b>Differences</b>		
<b>Item</b>	<b>Device</b>	<b>Predicate</b>
Intended Use	The Bio-Rad VARIANT II TURBO Link hemoglobin A1c Program	The Bio-Rad VARIANT II TURBO Hemoglobin A1c Program is intended

<b>Differences</b>		
<b>Item</b>	<b>Device</b>	<b>Predicate</b>
	is intended for the percent determination of hemoglobin A1c in human whole blood using ionexchange high-performance liquid chromatography (HPLC). The VARIANT II TURBO Link Hemoglobin A1c Program is for use with the VARIANT II TURBO Link Hemoglobin Testing System interfaced with an automated sample transport system. The Bio-Rad VARIANT II TURBO link Hemoglobin A1c Program is for Professional Use Only.	for the percent determination of hemoglobin A1c in human whole blood using ion-exchange high performance liquid chromatography (HPLC). The Bio-Rad VARIANT II TURBO Hemoglobin A1c Program is intended for Professional Use Only. For In Vitro Diagnostic Use.
Sample transport mode of operation	Continuous feed, batch or STAT mode of closed EDTA sample tubes from automated sample transport system.	Batch mode of closed EDTA sample tubes
Automated sample transport system	The VARIANT II TURBO Link Hemoglobin Testing System requires an external automated sample transport system [Sysmex HST-N (Hemoglobin Sample Transport) System].	The VARIANT II TURBO Hemoglobin Testing System is complete with an automated sample conveyor system.

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

Medical devices - Application of risk management to medical devices (ISO 14971:2000)

Medical Devices - Symbols to be used with medical device labels, labeling and

information to be supplied (ISO 15223)

Evaluation of the Linearity of Quantitative Measurement; Approved Guideline-Second Edition (CLSI EP6-A)

Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline-Second Edition (CLSI EP5-A2)

Draft Guidance Document for 510(k) Submission of Glycohemoglobin (Glycated or Glycosylated) Hemoglobin for IVDs (FDA guidance)

Guidance for Off-the-Shelf Software Use in Medical Devices; Final (FDA Guidance for Industry, FDA Reviewers and Compliance)

Guidance for the Content of Premarket Submissions for Software Contained in Medical Devices (FDA Guidance for Industry and FDA Staff)

Format for Traditional and Abbreviated 510(k)s – (FDA Guidance for Industry and FDA Staff)

#### **L. Test Principle:**

The VARIANT II TURBO Link Hemoglobin A1c Program is based on the chromatographic separation of HbA1c on a cation exchange cartridge. The various forms of hemoglobin exhibit charge differences (positive) at the acidic pH of the mobile phase, and thus can be separated on a support that is negatively charged (cation exchange). The use of ion-exchange chromatography then allows molecules to be separated based upon their charge. Separation is optimized to eliminate interferences from hemoglobin variants (HbS and HbC), labile A1c, hemoglobin F and carbamylated hemoglobin.

#### **M. Performance Characteristics (if/when applicable):**

##### **1. Analytical performance:**

###### ***a. Precision/Reproducibility:***

The first precision study was performed at Bio-Rad on the VARIANT II TURBO Link Hemoglobin A1c Program using EDTA whole blood patient samples tested with normal and diabetic hemoglobin A1c content. The method of precision analysis was performed using a protocol based on CLSI Evaluation Protocol, Vol.24, No. 25, EP5-A2 (2004) “Evaluation of Precision Performance of Clinical Chemistry Devices”. In this study, 40 runs (2 per day) were performed on one VARIANT II TURBO Link Hemoglobin Testing System over 20 working days. In each run, one aliquot of normal HbA1c and diabetic HbA1c patient samples, controls and four (4) patient samples used as filler to reproduce normal run

conditions were each analyzed in duplicate. The position of the precision specimens in each run was randomized.

	<b>Diabetes Control 1</b>	<b>Diabetes Control 2</b>	<b>Normal Patient (HbA1c)</b>	<b>Diabetic Patient (HbA1c)</b>
n= (number of samples)	80	80	80	80
Mean (HbA1c)	5.49	10.10	6.01	9.43
Within run (%CV)	0.96	0.41	0.62	0.47
Between day (%CV)	1.45	0.53	1.07	0.73
Between run (%CV)	0.00	0.76	0.26	0.29
Total Precision (%CV)	1.74	1.02	1.27	0.92

In the second study which was performed at a second site, two runs were performed each day for five days on one VARIANT II TURBO Link system. One normal and one diabetic patient were analyzed in duplicate in randomized order within each run. The results of the precision study are summarized in below.

	<b>Normal Sample (HbA1c)</b>	<b>Diabetic Sample (HbA1c)</b>
n= (number of samples)	40	40
Mean (HbA1c)	5.7	8.5
Within Run (%CV)	0.39	0.98
Between Day (%CV)	1.25	2.39
Between Run (%CV)	0.55	0.00
Total Precision (%CV)	1.43	2.58

*b. Linearity/assay reportable range:*

To demonstrate the linearity of the HbA1c measurement on the VARIANT II TURBO Link Hemoglobin A1c Program throughout the reportable range, low (4.1%) and high (17.6%), EDTA whole blood patient samples were assessed following CLSI EP6-A guideline “Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach”. The diabetic specimen was mixed with the normal specimen in varying ratios (i.e.3:1, 1:1 and 1:3) and the measured values were compared to the theoretical values based upon the dilution factor. Polynomial regression analysis (for first, second, and third order polynomials) was performed to determine the statistical significance of the non-linearity. These mixed samples were analyzed in duplicate on the VARIANT II TURBO Link Hemoglobin A1c Program using the VARIANT II TURBO Link Hemoglobin Testing System with Sysmex HST-N conveyor. Linear regression analysis calculated a slope of 1.0039, an intercept of 0.0541, and a correlation coefficient of 0.9996. The measured results for each dilution were within  $\pm 1.0\%$  of the predicted values.

<b>Sample Pool</b>	<b>Measured %A1c</b>	<b>Predicted %A1c</b>
1	4.10	4.16
2	7.60	7.68
3	10.93	11.02
4	14.47	14.58
5	17.57	17.69

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

Calibrators were previously cleared under k040872.

*d. Detection limit:*

The reportable range is 4.1 to 17.6% HbA1c (see linearity section above)

*e. Analytical specificity:*

Interfering substance studies performed included labile A1c, carbamylated hemoglobin and hemoglobin F. Studies such as bilirubin, lipemia and hemoglobin S and C were filed with the VARIANT II TURBO Hemoglobin A1c Program k040872. These studies were not repeated because the reagents are the same.

Labile A1c concentrations up to 4.7% in non-diabetic patients (4.27-6.0% HbA1c) and up to 6.4% in diabetic patients do not interfere with the assay.

Hemoglobin F concentrations up to 5% do not interfere with the assay.

Carbamylated hemoglobin concentrations up to 3.1% in non-diabetic patients (4.27- 6.07% HbA1c) and up to 5.5% in diabetic patients do not interfere with the assay.

To test the level of interference of carbamylated hemoglobin on the VARIANT II TURBO Link Hemoglobin A1c Program, patient EDTA whole blood specimen pools with normal and diabetic levels of A1c were split into two aliquots. The Red Blood Cells (RBCs) of both aliquots were washed with phosphate buffered saline (PBS). One aliquot of each level was incubated with a 0.05 M potassium cyanate solution until the carbamylated hemoglobin level increased by approximately 1, 2, and 3%. Aliquots from each level were analyzed in duplicate on the VARIANT II TURBO Link Hemoglobin A1c Program on one VARIANT II TURBO Link Hemoglobin Testing System with Sysmex HST-N conveyor. The specification for interference states that the difference between a normal control (without spiked CHb) and a normal test sample (with spiked CHb) with up to 1.5 % CHb is not greater than + 0.3% HbA1c and the difference between a diabetic control (without spiked CHb) and a diabetic test sample (with spiked CHb) with up to 1.5% CHb is no greater than + 0.5% HbA1c. Carbamylated hemoglobin concentrations up to 3.1% in non-diabetic patients (4.27 – 6.07% HbA1c) and up to 5.5% in diabetic patients did not interfere with the assay.

To determine if HbF interferes with HbA1c quantitation on the VARIANT IITURBO Link Hemoglobin A1c Program, two pooled EDTA whole blood patient samples representing normal and diabetic HbA1c levels were split into two aliquots. One aliquot was spiked with approximately 3, 5, 10 and 15% purified HbF. The samples were run on a VARIANT II TURBO Link Hemoglobin Testing System with Sysmex HST-N conveyor. The difference in %A1c value between a normal control (without spiked HbF) and a normal test sample (with spiked HbF) had to be < 0.3% to be defined as showing no interference. The difference in %A1c value between the diabetic control (without spiked HbF) and diabetic test sample (with spiked HbF) had to be < 0.5% to be defined as showing no interference. The results indicated that HbF levels up to 5% have no significant effect on HbA1c determination.

To study the interference from labile A1c on the VARIANT II TURBO Link Hemoglobin A1c Program, two EDTA whole blood patients samples representing normal and diabetic A1c levels were split into aliquots. These aliquots were supplemented by the addition of an aqueous glucose stock solution. The samples were incubated for three hours at 37°C to facilitate formation of labile A1c. The samples were then run on a VARIANT II TURBO Link Hemoglobin Testing System with Sysmex HST-N conveyor, and the samples that were closest to 1, 2, and 3% labile A1c were selected. The specification for no interference is that the difference in HbA1c value between a control and test sample with up to 3% labile A1c is no greater than + 0.3% HbA1c. The results indicate that the difference in HbA1c was 0.2%



between spiked samples and the control for the normal sample. There was no difference between spiked samples and the control sample for the diabetic sample. Labile A1c concentrations up to 4.7% in non-diabetic patients (4.27 – 6.07% HbA1c) and up to 6.4% in diabetic patients do not interfere with the assay.

*f. Assay cut-off:*

Not applicable

## 2. Comparison studies:

### *a. Method comparison with predicate device:*

To demonstrate accuracy across the measuring range of 4.1 to 17.6 % A1c, the VARIANT II TURBO Link Hemoglobin A1c Program was compared to the predicate VARIANT II TURBO Hemoglobin A1c Program. 22 EDTA whole blood patient samples and 29 spiked EDTA whole blood samples were run in singlicate on the VARIANT II TURBO Link Hemoglobin A1c Program and the VARIANT II TURBO Hemoglobin A1c Program. The range of samples run on the VARIANT II TURBO Link Hemoglobin A1c Program was 3.9 to 17.3 % A1c. A linear regression analysis of the results calculated a slope of 0.961, an intercept of 0.303, and a correlation coefficient of 0.999.

A second study performed at a current customer site evaluated the VARIANT II TURBO Link Hemoglobin A1c Program compared to the VARIANT II TURBO Hemoglobin A1c Program. A comparison of 180 EDTA whole blood patient samples with values distributed from 4.2 to 14.0 % HbA1c were run in singlicate on both systems. A linear regression analysis of these results calculated a slope of 0.983, an intercept of 0.225, and a correlation coefficient of 0.998.

### *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:

Hemoglobin A1c Expected Value Range was determined from literature (American Diabetes Association. Standards of Medical Care for Patients with Diabetes Mellitus. Diabetes Care 2001, 24 (Suppl. 1), 33-43). Hemoglobin A1c > 8% Action is suggested. Hemoglobin A1c < 7% is the goal. Hemoglobin A1c < 6% is non-diabetic level.

**N. Instrument Name:**

VARIANT II TURBO Link Hemoglobin Testing System

**O. System Descriptions:**

1. Modes of Operation:

The VARIANT II TURBO Link Hemoglobin Testing System in combination with an automated transport system, such as the Sysmex HST-N System, can run a closed tube in different modes. It can run a STAT sample, batch of samples or continuous feed of samples

2. Software:

The VARIANT II TURBO Link Hemoglobin Testing System incorporates a dedicated software package, called CDM 4.1, of instrument control, data control, results analysis, calibration, quality control and service software. Software documentation has been included in this submission for CDM 4.1

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes   X   or No           

3. Specimen Identification:

Samples are identified on the VARIANT II TURBO Link Hemoglobin Testing System by barcodes on each primary sample tube, corresponding to the sample's accession identification. Non-patient samples (microvials with controls, calibrators, primer, etc) have specific barcodes that indicate sample type (e.g. Cal 1 for Calibrator 1).

As a rack moves toward the VARIANT II TURBO Link VSS (VARIANT

Sampling Station), the barcode reader attempts to read the tube barcode. If the barcode is successfully read, and there is an A1c order for the sample, then the barcode, rack, and rack position are provided to the VARIANT II TURBO Link VSS. The VARIANT II TURBO Link VSS provides this information to the CDM 4.1 software, which inserts the information into a Worklist of samples and links the results to this particular identifier.

If the barcode is missing or cannot be read, the sample can either be skipped (assume an A1c order is absent) or processed (assume an A1c order is present). If the sample is processed, then a sequential ERR barcode number is generated, along with the rack and rack position for the sample. It is then handled by the VSS and CDM as noted above.

4. Specimen Sampling and Handling:

The Instructions for Use for the VARIANT II TURBO Link Hemoglobin A1c Program instruct the user that there is no sample preparation necessary, except for abnormal size tubes. The Primary sample tubes (EDTA whole blood) are loaded into the Sysmex HST-N sample racks and placed on the Sysmex HST-N transport line. For sample identification, it is necessary to use adapters (provided by Bio-Rad) when processing microvials for use of prediluted samples. Microvials with pierceable caps are used to prevent spillage.

5. Calibration:

Calibration is performed at regular intervals. For each VARIANT II TURBO Link Hemoglobin A1c Program, Bio-Rad provides calibrator sets of known analyte concentrations. New analytical or guard cartridges require two levels of calibration before patients or controls can be processed to provide concentrations of percent hemoglobin A1c.

6. Quality Control:

External Quality Control materials must be purchased separately. In keeping with good laboratory practice, diabetic and non-diabetic control specimens should be included in each run. A repeat run is indicated when expected control values are not obtained.

**P. Other Supportive Instrument Performance Characteristics Data Not Covered In The “Performance Characteristics” Section above:**

Not applicable

**Q. Proposed Labeling:**

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

**R. Conclusion:**

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