DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
Document Control Center – WO66-G609
Silver Spring, MD 20993-0002

BIO-RAD LABORATORIES, INC.
JACKIE BUCKLEY
REGULATORY AFFAIRS REP IV
4000 ALFRED NOBEL DR.
HERCULES CA 94547

December 9, 2015

Re: K151321
Trade/Device Name: D-100™ HbA1c
D-100™ HbA1c Calibrator Pack
Regulation Number: 21 CFR 862.1373
Regulation Name: Glycosylated hemoglobin assay
Regulatory Class: II
Product Code: PDJ, LCP, JIT
Dated: November 06, 2015
Received: November 09, 2015

Dear Jackie Buckley:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820); and if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

If you desire specific advice for your device on our labeling regulations (21 CFR Parts 801 and 809), please contact the Division of Industry and Consumer Education at its toll-free number (800) 638 2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Industry and Consumer Education at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm.

Sincerely yours,

Katherine Serrano -S

For: Courtney H. Lias, Ph.D.
Director
Division of Chemistry and Toxicology Devices
Office of In Vitro Diagnostics
and Radiological Health
Center for Devices and Radiological Health

Enclosure

DEPARTMENT OF HEALTH AND HUMAN SERVICES
Food and Drug Administration

Indications for Use

Form Approved: OMB No. 0910-0120
Expiration Date: January 31, 2017
See PRA Statement below.

510(k) Number *(if known)*
K151321

Device Name
D-100™ HbA1c
D-100™ HbA1c Calibrator Pack

Indications for Use *(Describe)*
The D-100™ HbA1c test is intended for the quantitative determination of hemoglobin A1c (IFCC mmol/mol and NGSP %) in human whole blood using ion-exchange high-performance liquid chromatography (HPLC) on the D-100 Hemoglobin Testing System.

Hemoglobin A1c measurements are used as an aid in diagnosis of diabetes mellitus, as an aid to identify patients who may be at risk for developing diabetes mellitus, and for the monitoring of long-term blood glucose control in individuals with diabetes mellitus.

The D-100™ HbA1c test is intended for Professional Use Only.

Calibrators:
The D-100™ HbA1c Calibrator Pack is for the calibration of the D-100 Hemoglobin Testing System used for the quantitative determination of hemoglobin A1c(HbA1c) in human whole blood.

Type of Use *(Select one or both, as applicable)*

☒ Prescription Use (Part 21 CFR 801 Subpart D) ☐ Over-The-Counter Use (21 CFR 801 Subpart C)

CONTINUE ON A SEPARATE PAGE IF NEEDED.

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510(k) Summary (Summary of Safety and Effectiveness)

This Summary of 510(k) Safety and Effectiveness is being submitted in accordance with the requirements of SMDA 1990 and 21 CFR 807.92.

The assigned 510(k) number is: K151321_____.

Date Summary prepared: Nov. 4, 2015

1. **Applicant Name:**
   Bio-Rad Laboratories, Inc.
   Clinical Diagnostics Group
   4000 Alfred Nobel Drive
   Hercules, California 94547

2. **Contact Person(s):**
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   E-Mail:al_evans@bio-rad.com

3. **Device Name/Trade Name:**
   Reagents:
   Trade Name: D-100™ HbA1c
   Classification Name: Assay, Glycosylated Hemoglobin
   Common Name: HbA1c
   Product Code: PDJ, LCP
   C.F.R Section: 21 CFR 862.1373
   Device classification: Class II
   Panel Classification: Chemistry

   Calibrators:
   Trade Name: D-100™ HbA1c Calibrator Pack
   Classification Name: Calibrator, Secondary
   Common Name: Calibrator
   Product Code: JIT
   C.F.R Section: 21 CFR 862.1150
   Device classification: Class II
   Panel Classification: Clinical Chemistry

4. **Predicate Device:**

Predicate Device Name	**Predicate Device 510(k) Number**
VARIANT II TURBO HbA1c Kit -2.0	K142448
VARIANT II Hemoglobin A1c Calibrators	K070452

5. Description of the Device:

The Bio-Rad D-100™ HbA1c utilizes principles of ion-exchange high-performance liquid chromatography (HPLC). A high-pressure pumping system delivers a buffer solution to an analytical cartridge and detector. Whole blood samples undergo an automatic hemolysis and dilution process before being introduced into the analytical flow path. Prediluted samples are identified based upon the use of a microvial adapter in the sample rack, and the automatic dilution step is omitted.

A programmed buffer gradient of increasing ionic strength delivers the sample to the analytical cartridge where the hemoglobin species are separated based upon their ionic interactions with the cartridge material and the buffer gradient. The separated hemoglobin species then pass through the flow cell where changes in the absorbance are measured at 415 nm and recorded as a digital chromatogram.

The software performs an analysis of the hemoglobin peaks in the chromatogram, recording information including retention time, peak area, and relative are percent. Any peaks that are identified as the target analyte(s) are calibrated before generating a sample report and chromatogram for each sample. The software includes an optional feature (Advisor) that compares the sample report against a set of rules that are programmed to take user-specified actions.

The D-100™ HbA1c test is designed to be used on the D-100™ Hemoglobin Testing System.

Reagents:
The D-100™ HbA1c reagents contain the following components:

Description
D-100 ™ HbA_{1c} Analytical Cartridge/Calibrator Pack. One pack consisting of: ▪ Cation exchange cartridge. 10,000 tests each ▪ Calibrator Pack: 1 vial of Conditioner, 1 vial of Calibrator Level 1, and 1 vial of Calibrator Level 2. The vials contain lyophilized human whole blood with glycine and trehalose as preservatives.
D-100 ™ Prefilters. 2000 tests each. Package of 5.
D-100 ™ Cleaning Tube. One microvial containing 1.5 mL of a liquid cleaning solution. Single use.
D-100 ™ Sample Diluent. Each bottle contains 1 L of deionized water with <0.05% sodium azide as a preservative.
D-100 ™ HbA_{1c} Elution Buffer A. Each bottle contains 2600 mL of a succinate/sodium perchlorate buffer. Contains <0.05% sodium azide as a preservative.
D-100 ™ HbA_{1c} Elution Buffer B. Each bottle contains 1400 mL of a succinate/sodium perchlorate buffer. Contains<0.05% sodium azide as a preservative.

D-100 ™ Wash Solution. Each bottle contains 3300 mL of deionized water with <0.05% sodium azide as a preservative.

Calibrator:
Each Calibrator Pack contains Calibrator values which have been value assigned using secondary calibrators that are traceable to the International federation of Clinical Chemistry and Laboratory Medicine (IFCC) reference method.

Description
D-100 ™ HbA_{1c} Calibrator Pack. One pack consisting of 1 vial of Conditioner, 1 vial of Calibrator Level 1, and 1 vial of Calibrator Level 2. The vials contain lyophilized human whole blood with glycine and trehalose as preservatives.

6. Indications for Use:

The D-100™ HbA1c test is intended for the quantitative determination of hemoglobin A1c (IFCC mmol/mol and NGSP %) in human whole blood using ion-exchange high-performance liquid chromatography (HPLC) on the D-100™ Hemoglobin Testing System.

Hemoglobin A1c measurements are used as an aid in diagnosis of diabetes, as an aid to identify patients who may be at risk for developing diabetes mellitus, and for the monitoring of long-term blood glucose control in individuals with diabetes mellitus.

The Bio-Rad D-100™ HbA1c test is intended for Professional Use Only.

Calibrators
The D-100 HbA1c Calibrator Pack is for the calibration of the D-100 Hemoglobin Testing System used for the quantitative determination of hemoglobin A1c (HbA1c) in human whole blood.

7. Substantial Equivalence Information:

Predicate Device Information:

Predicate Device Name	**Predicate Device 510(k) Number**
VARIANT II TURBO HbA1c Kit -2.0	K142448
VARIANT II Hemoglobin A1c Calibrators	K070452

The comparison of the technological characterizes of the D-100 HbA1c assay (candidate assay) utilizes principles of ion-exchange high-performance liquid chromatography (HPLC) similar to the same technology of the VARIANT II TURBO HbA1c Kit – 2.0 (predicate device).
Tables 1 and 2 provide the similarities and differences between the candidate assay and the predicate assay.

Table 1: Reagent Similarities and Differences

Reagent **Similarities and Differences**		
Features	**Candidate Device:** **D-100™ HbA1c** (K)151321	**Predicate Device:** **VARIANT™ II TURBO HbA1c Kit – 2.0** (K)142448
Intended Use	Same	Intended for the quantitative determination of hemoglobin A1c (IFCC mmol/mol and NGSP %)
Platform	D-100™ Hemoglobin Testing System	VARIANT™TURBO Hemoglobin Testing System and VARIANT™TURBO Link Hemoglobin Testing System
Measuring Interval	3.5 to 20% (NSGP) 15 – 195 mmol/mol HbA1c (IFCC)	3.4 to 20.6 % (NSGP) 14 – 203 mmol/mol HbA1c (IFCC)
Specimen Type	Same	Human Whole blood
Assay Principle	Same	Ion exchange HPLC
Matrices	K_2-EDTA, K_3-EDTA Potassium Oxalate/Sodium Fluoride, Sodium Citrate, Sodium Heparin, Lithium Heparin	K_2-EDTA, K_3-EDTA Hemoglobin Capillary Collection Kit
Standardization	Same	Traceable to the Diabetes Control and Complications Trial (DCCT) reference method and IFCC. Certified via the National Glycohemoglobin Standardization Program (NGSP)

Table 2: Calibrator Similarities and Differences

Calibrator Similarities and Differences		
Features	**Candidate Device: D-100™ HbA1c Calibrator Pack** (K)151321	**Predicate Device: VARIANT™ II Hemoglobin A1c Calibrators** (K) 070452
Intended Use	Same	Intended for the quantitative determination of hemoglobin A1c in Human Whole Blood
Levels	Same	Levels 1 & 2 Calibration is performed once at the beginning of a new cartridge.
Standardization/Traceability	Same	Each lot of calibrators is value assigned and values are reported in both NGSP and IFCC units.

8. Summary of Nonclinical Performance Data:

a. Precision/Reproducibility:

The precision of the D-100™ HbA1c test was evaluated based on CLSI EP05-A2 guidelines, Evaluation of Precision Performance of Quantitative Measurement Methods using a modified study design. Four EDTA whole blood samples at the following targeted HbA1c concentrations of ~5%, ~6.5%, ~8% and ~12% were utilized in the study. In addition, five quality control materials were also tested. Precision was evaluated using three reagent lots, three D-100™ I Hemoglobin Testing Systems at two different sites. The samples were run in duplicate in 2 runs per day for 20 day.
NGSP results are shown in Tables 3-6. IFCC results are shown in Tables 7-10.

Table 3: Instrument 1 (% CV by Sample (NGSP)

Variation Source	Instrument ID: SM93								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP%)	5.5%	9.4%	5.2%	6.7%	8.1%	12.0%	5.3%	9.9%	14.8%
Repeatability	0.7%	0.7%	0.7%	0.7%	0.7%	0.6%	0.8%	0.7%	0.7%
Between-Run	0.0%	0.1%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Day	0.6%	0.2%	0.3%	0.2%	0.1%	0.3%	0.3%	0.1%	0.2%
Between-Lot	1.2%	0.8%	1.3%	1.1%	1.0%	0.6%	1.4%	0.8%	0.6%
Total Precision	1.5%	1.1%	1.5%	1.3%	1.2%	0.9%	1.6%	1.0%	0.9%

Table 4: Instrument 2 (% CV by Sample (NGSP)

Variation Source	Instrument ID: SM94								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP%)	5.5%	9.4%	5.2%	6.6%	8.1%	12.0%	5.3%	10.0%	14.8%
Repeatability	1.1%	0.9%	0.8%	0.9%	1.0%	0.9%	1.0%	1.0%	1.0%
Between-Run	0.0%	0.3%	0.0%	0.0%	0.2%	0.0%	0.0%	0.1%	0.1%
Between-Day	0.6%	0.2%	0.5%	0.5%	0.3%	0.3%	0.2%	0.4%	0.3%
Between-Lot	1.2%	0.2%	1.5%	0.6%	0.0%	0.3%	1.5%	0.1%	0.5%
Total Precision	1.7%	1.0%	1.8%	1.2%	1.0%	1.0%	1.8%	1.1%	1.1%

Table 5: Instrument 3 (% CV by Sample (NGSP))

Variation Source	Instrument ID: SM98								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP%)	5.4%	9.4%	5.1%	6.6%	8.1%	12.0%	5.3%	9.9%	14.7%
Repeatability	1.0%	1.0%	1.0%	1.1%	0.9%	0.9%	1.0%	0.9%	0.8%
Between-Run	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Day	0.6%	0.5%	0.4%	0.5%	0.5%	0.4%	0.4%	0.4%	0.4%
Between-Lot	1.6%	1.0%	1.5%	1.5%	1.2%	1.1%	1.7%	1.0%	1.1%
Total Precision	1.9%	1.4%	1.9%	2.0%	1.6%	1.5%	2.0%	1.4%	1.4%

Table 6: Instruments Combined (% CV by Sample (NGSP))

Variation Source	All Instruments								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP %)	5.5%	9.4%	5.2%	6.6%	8.1%	12.0%	5.3%	9.9%	14.8%
Repeatability	0.9%	0.9%	0.9%	0.9%	0.9%	0.8%	0.9%	0.9%	0.8%
Between-Run	0.0%	0.0%	0.0%	0.0%	0.1%	0.0%	0.0%	0.0%	0.0%
Between-Day	0.6%	0.3%	0.4%	0.4%	0.4%	0.3%	0.3%	0.3%	0.3%
Between-Instrument	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Lot	1.4%	0.7%	1.5%	1.1%	0.9%	0.7%	1.5%	0.7%	0.8%
Total Precision	1.7%	1.2%	1.7%	1.5%	1.3%	1.2%	1.8%	1.2%	1.2%

Table 7: Instrument 1 (% CV by Sample (IFCC Units- mmol/mol))

Variation Source	Instrument ID:SM93								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (IFCC)	36.5	79.3	33.2	49.4	65.4	108.1	34.5	85.0	137.7
Repeatability	1.2%	0.9%	1.2%	1.0%	0.9%	0.8%	1.3%	0.8%	0.8%
Between-Run	0.0%	0.1%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Day	0.9%	0.3%	0.5%	0.4%	0.2%	0.4%	0.6%	0.2%	0.3%
Between-Lot	2.0%	1.1%	2.2%	1.6%	1.3%	0.7%	2.3%	1.0%	0.7%
Total Precision	2.5%	1.4%	2.5%	1.9%	1.6%	1.1%	2.7%	1.3%	1.1%

Table 8: Instrument 2 (% CV by Sample (IFCC Units- mmol/mol))

Variation Source	Instrument ID: SM94								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP%)	36.4	79.2	32.9	49.0	65.1	108.0	34.5	85.4	137.9
Repeatability	1.7%	1.2%	1.4%	1.4%	1.3%	1.1%	1.7%	1.3%	1.1%
Between-Run	0.0%	0.4%	0.0%	0.0%	0.3%	0.0%	0.0%	0.1%	0.1%
Between-Day	1.0%	0.3%	0.8%	0.7%	0.4%	0.4%	0.3%	0.5%	0.4%
Between-Lot	2.1%	0.3%	2.6%	1.0%	0.0%	0.4%	2.5%	0.1%	0.6%
Total Precision	2.9%	1.4%	3.1%	1.8%	1.4%	1.2%	3.1%	1.4%	1.3%

Table 9: Instrument 3 (% CV by Sample (IFCC Units- mmol/mol)

Variation Source	Instrument ID: SM95								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP%)	36.0	79.1	32.7	48.9	64.9	107.7	34.1	84.8	137.6
Repeatability	1.6%	1.2%	1.8%	1.7%	1.3%	1.1%	1.6%	1.2%	1.0%
Between-Run	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Day	1.0%	0.6%	0.8%	0.8%	0.7%	0.5%	0.7%	0.5%	0.5%
Between-Lot	2.6%	1.2%	2.6%	2.3%	1.6%	1.3%	2.9%	1.2%	1.2%
Total Precision	3.2%	1.8%	3.3%	2.9%	2.1%	1.8%	3.4%	1.7%	1.7%

Table 10: Instruments Combined (% CV by Sample (IFCC Units- mmol/mol)

Variation Source	All Instruments								
	Control 1	Control 2	Patient 1	Patient 2	Patient 3	Patient 4	QC 1	QC 2	QC 3
Concentration HbA1c (NGSP %)	36.3	79.2	33.0	49.1	65.1	107.9	34.3	85.1	137.8
Repeatability	1.5%	1.1%	1.5%	1.4%	1.2%	1.0%	1.5%	1.1%	1.0%
Between-Run	0.0%	0.0%	0.0%	0.0%	0.1%	0.0%	0.0%	0.0%	0.0%
Between-Day	1.0%	0.4%	0.7%	0.6%	0.5%	0.4%	0.6%	0.4%	0.4%
Between-Instrument	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Between-Lot	2.2%	1.0%	2.5%	1.7%	1.2%	0.9%	2.6%	0.9%	0.9%
Total Precision	2.9%	1.5%	3.0%	2.3%	1.7%	1.4%	3.1%	1.5%	1.4%

b. Linearity

A linearity study was performed per CLSI EP06-A: Evaluation of the Linearity of Quantitative Measuring Procedures; A Statistical Approach. Linearity across the reportable range was performed using low (3.5%HbA1c) and high (20%HbA1c) EDTA whole blood patient samples. These samples were mixed together in varying ratios. The measured values were compared to the theoretical values based upon the dilution factor. Polynomial regression analysis (for first, second, and third order polynomials) were performed to determine the statistical significance of non-linearity. The higher order coefficients were found not to be significant and linearity was demonstrated.

% HbA1c (NGSP) using the D-100™ HbA1c test has been demonstrated linear from 3.5 – 20.0% HbA1c with the maximum measured difference of

± 0.09% between the predicted 1st and 2rd order results as shown in Table 11 below. Mmol/mol HbA1c (IFCC) has been demonstrated as linear from 15 – 195 mmol/mol with the maximum measured difference of ± 0.9% (or +/- 0.94mmol/mol) as shown in Table 12 below.

Table 11: Results of Linearity Study (NGSP %)

Sample Pool	Predicted 1st Order	Predicted 2nd Order	Difference
Control, Level 1	3.50	3.43	0.07
Level 2	5.16	5.14	0.02
Level 3	6.82	6.85	-0.03
Level 4	8.48	8.54	-0.06
Level 5	10.13	10.21	-0.08
Level 6	11.79	11.88	-0.09
Level 7	13.44	13.53	-0.09
Level 8	15.09	15.17	-0.08
Level 9	16.74	16.79	-0.05
Level 10	18.39	18.41	-0.02
High, Level 11	20.04	20.01	0.03

Table 12: Results of Linearity Study (IFCC mmol/mol)

Sample Pool	Predicted 1st Order	Predicted 2nd Order	Difference
Control, Level 1	14.78	13.85	0.92
Level 2	32.93	32.56	0.37
Level 3	51.07	51.12	-0.28
Level 4	69.19	69.55	-0.36
Level 5	87.30	87.83	-0.54
Level 6	105.38	105.98	-0.60
Level 7	123.46	123.99	-0.53
Level 8	141.51	141.86	-0.34
Level 9	159.55	159.59	-0.04
Level 10	177.57	177.18	0.39
High, Level 11	195.58	194.63	0.94

c. Method Comparison

A Method comparison study was performed per CLSI EP09-A2 IR, Method Comparison and Bias Estimation Using Patient Samples. 129 variant-free whole blood EDTA samples ranging from 3.5% to 20.0% HbA1c were evaluated using the D-100™ HbA1c on the D-100™ Hemoglobin Testing System. Samples were tested in a single determination over a 4 day period. The results were compared to testing performed at a secondary NGSP SRL reference laboratory using a cleared HPLC-based HbA1c assay. The distribution of samples spanned the measuring interval listed in Table 13.

Table 13: Distribution of samples

Hemoglobin A1c level	n	% Samples tested
≤ 5%	6	4.7
5 – 6%	17	13.2
6 – 6.5%	31	24.0
6.5 – 7%	33	25.6
7 – 8%	20	15.5
8 – 9%	11	8.5
> 9%	11	8.5
Total samples	129	100

Deming (weighted) and Passing-Bablok regression analyses were performed for the D-100™ HbA1c versus the NGSP SRL reference method. Deming (weighted), Passing-

Bablok and Linear regression analyses were performed for the D-100™ HbA1c on the D-100 Hemoglobin Testing System versus the reference G8 HPLC method are summarized in Table 14.

Table 14: Summary of Method Comparison Results

	y-Intercept	95% CI	Slope	95% CI
Deming	0.0223	-0.0684 - 0.1131	0.9867	0.9736 – 0.9999
Passing-Bablok	-0.0091	-0.0803 – 0.0763	0.9909	0.9789 – 1.0026

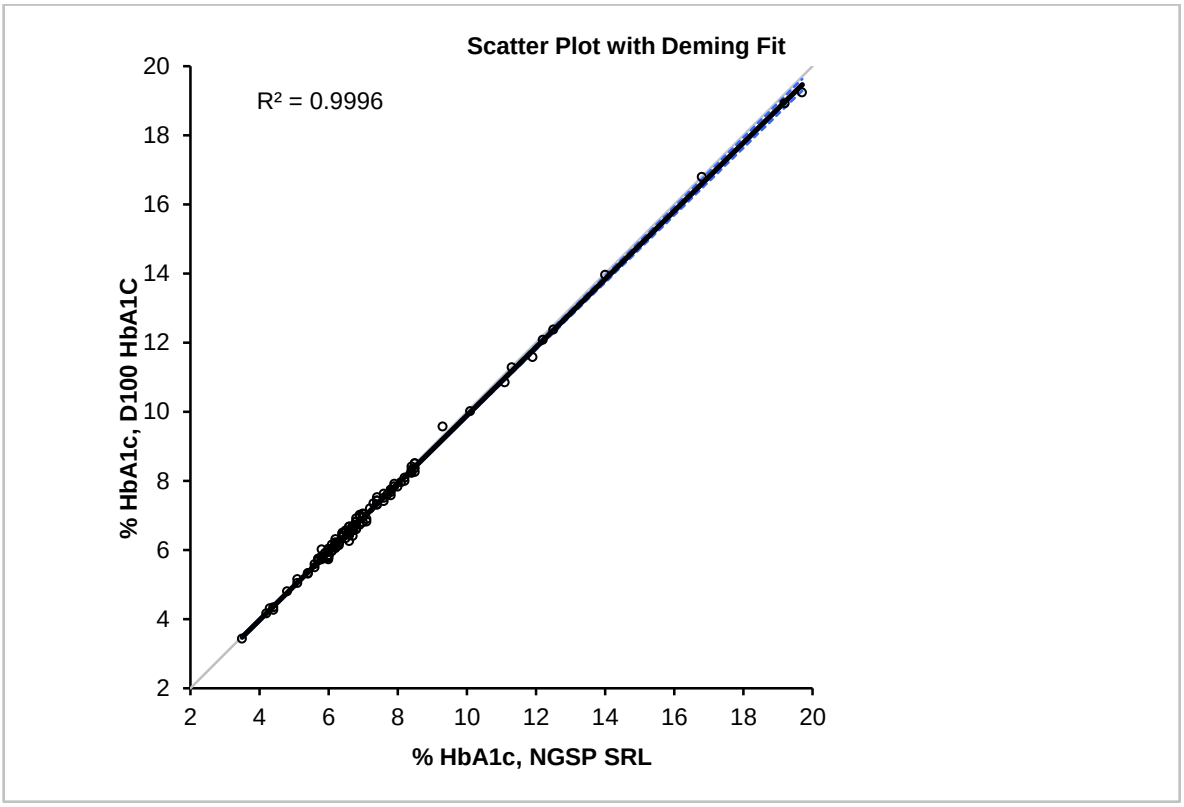

Figure 1: Scatter Plot using Deming Fit, %HbA1c, NGSP SRL vs. D-100 HbA1c.

(1) The following biases between D-100 HbA1c versus NGSP SRL Method (Reference method) were observed in Table 15.

Table 15: Bias Estimation

% HbA1c – Decision Level	Bias	% Bias
5.0	-0.047	-0.85

6.5	-0.066	-0.98
8.0	-0.090	-1.11
12.0	-0.190	-1.57

Total Error Decision Levels

Using the results of bias estimation (%Bias) in the method comparison study and precision estimates in the reproducibility study, Total Error (TE) at four concentrations: (5.0 %, 6.5%, 8.0% and 12.0%) were calculated as follows: %TE=|%Bias| + 1.96 *CV* (1 + %Bias). The results are presented in Table 16.

Table 16: Total Error Estimation

% A1c – Decision Level	% Bias	% CV	% TE
5.0	-0.85	1.7	4.2
6.5	-0.98	1.5	3.9
8.0	-1.11	1.3	3.6
12.0	-1.57	1.2	3.9

d. Traceability, Stability, Expected Values (calibrators)

The D-100 HbA1c test standardization is traceable to the International Federation of Clinical Chemistry (IFCC) reference calibrators. The D-100 HbA1c assay is NGSP certified. The NGSP certification expires in one year. See NGSP website for current certification at http://www.ngsp.org. The derived results of (%) from the NGSP correlation are calculated from the individual quantitative results for Hemoglobin A1c (HbA1c). The International Federation of Clinical Chemistry (IFCC) units of mmol/mol are calculated using the Master Equation NGSP (%) = 0.09148 x IFCC (mmol/mol) + 2.152. HbA1c results are provided to the customers using two different units: NGSP equivalent units (%) and IFCC equivalent units (mmol/mol).

Calibrator Materials:
Value assignment for D-100™ HbA1c Calibrators are traceable to IFCC reference method and can be transferred to DCCT/NGSP by calculation.

Stability:
Shelf life claims: Un-opened calibrators can be stored at 2-8°C until the expiration date or for 24 months.
Open-vial claims: The recommended storage condition for in-use calibrators is one day. On-board stability for the D-100 HbA1c calibrator pack and reagents demonstrated 90 days stability on the D-100 Hemoglobin Testing System.

e. Analytical specificity:

i.) Endogenous Interference

An Endogenous Interference study was performed per CLSI EP07-A2, Interference Testing in Clinical Chemistry. Two EDTA whole blood sample pools were evaluated using a low level whole blood sample with a concentration ~6.5%HbA1c and a high level whole blood sample with a concentration of HbA1c of ~8.0%.

Conjugated bilirubin, unconjugated bilirubin and glucose, available in pure

form, were obtained and stock solutions prepared at 10x the intended test concentration. The 10x stock solution of the test substance was pipetted into a low whole blood sample pool (at ~6.5% HbA1c) and a high whole blood sample pool (~8.0% HbA1c), making the test pool. Ten replicates of each pool prepared with the test and control samples were analyzed using the D-100™ HbA1c on the D-100™Hemoglobin Testing System.

Rheumatoid factor, lipemia and total protein were not available as pure standards therefore serum samples with known concentration of these compounds were used. The test pool was prepared by mixing the serum sample known to have a high test substance concentration with a whole blood non-variant sample such that the concentration of test substance in the final mixture would be at the desired level. Ten replicates of each pool prepared with the test and control samples were analyzed using the D-100™ HbA1c on the D-100™ Hemoglobin Testing System.

Significant interference was defined as a ± 7% change in %HbA1c value from the control. Results in Table 17 showed no significant interference up to the stated concentrations.

Table 17: Endogenous Interference Study Results

	Concentration	
Endogenous substance	**Conventional (US) units**	**SI Units**
Lipemia (Intralipid)	6000 mg/dL	60 g/L
Conjugated bilirubin	60 mg/dL	712 µmol/L
Unconjugated bilirubin	60 mg/dL	1026 µmol/L
Glucose	2000 mg/dL	111 mmol/L
Rheumatoid factor	750 IU/mL	750 kIU/mL
Total protein	21 g/dL	210 g/L

ii.) Drug Interference:

A Drug Interference study was performed based per CLSI EP07-A2, Interference Testing in Clinical Chemistry. Two EDTA whole blood sample pools were evaluated using a low level whole blood sample with a concentration ~6.5%HbA1c and a high level whole blood sample with a concentration of ~8.0%HbA1c. Test samples were prepared by spiking each drug at the interferent concentration shown in Table 18. Ten replicates of each drug prepared with the test and control samples were analyzed using the D-100™ HbA1c on the D-100™ Hemoglobin Testing System.

Significant interference was defined as a more than ± 7% change in %HbA1c value from the control. No significant interference was observed at therapeutic levels up to the stated concentrations in Table 18 on the following page.

Table 18: Drug Interference Study Results

Potential Drug Interferent	Highest Level Tested showing no Significant Interference	
	Conventional (US) units	SI units
Acetylcysteine	166 mg/dL	10.2 mmol/L
Ampicillin-Na	1000 mg/dL	28.65 mmol/L
Ascorbic acid	300 mg/dL	17.05 mmol/L
Cefoxitin	2500 mg/dL	58.55 mmol/L
Heparin	5000 U/L	5000 U/L
Levodopa	20 mg/dL	1015 µmol/L
Methyldopa	20 mg/dL	948 µmol/L
Metronidazole	200 mg/dL	11.7 mmol/L
Doxycyclin	50 mg/dL	1124 µmol/L
Acetylsalicylic acid	1000 mg/dL	55.51 mmol/L
Rifampicin	64 mg/L	78 µmol/L
Cyclosporine	5 mg/L	4 µmol/L
Acetaminophen	200 mg/L	1323 µmol/L
Ibuprofen	500 mg/L	2427 µmol/L
Theophylline	100 mg/L	556 µmol/L
Phenylbutazone	400 mg/L	1299 µmol/L

iii.) Cross Reactivity with Hemoglobin Derivatives:

A Hemoglobin Derivatives Interference study was performed based on CLSI EP07-A2, Interference Testing in Clinical Chemistry. Potential interference from Acetylated hemoglobin (Hb), Carbamylated hemoglobin (Hb) and Labile HbA1c were evaluated using a low level whole blood EDTA sample with a concentration ~6.5%HbA1c and a high level whole blood EDTA sample with a concentration of

~8.0% HbA1c. The potentially interfering hemoglobin derivatives were spiked into the low and high level blood samples and each sample was analyzed using ten replicates each in the same analytical run on the D-100™ Hemoglobin Testing System with the D-100™HbA1c.

Significant interference was defined as more than a ±7% change in HbA1c value from the control. The test result conclusions are as follows:

- Acetylated Hb- up to 50 mg/dL does not interfere with this assay.
- Carbamylated Hb – up to 5% does not interfere with this assay.
- Labile A1c- up to 1200 mg/dL of glucose does not interfere with this assay.

Results showed there was no cross reactivity with these substances at physiological levels.

iv.) Hemoglobin Variant Study:

A Hemoglobin Variant study was performed using specific variant samples known to contain hemoglobin variants S, C, E, D, A2 and F. Two whole blood EDTA patient samples containing an HbA1c ~6.5% and ~ 8% and the appropriate hemoglobin variant were tested. Testing of the samples containing hemoglobin variants S, C, E, D , A2 and F were performed in duplicate. Testing of the samples was performed using the D-100™ HbA1c on the D-100™ Hemoglobin Testing System and compared to results obtained by a NGSP reference method that has been demonstrated to be free from the hemoglobin interferent. Table 19 contains the number of samples, range of samples and concentration of samples used in the Hemoglobin Variant Study. Table 20 contains the results for the Hemoglobin Variant study bias.

Table 19: Variant samples used in Hemoglobin Variant Study

Hemoglobin Variant	n	Range in % Abnormal Variant	Range in %HbA1c Concentration
HbS	20	28.7 – 40.2	5.6 – 9.6
HbC	20	34.4 – 44.1	5.0 – 10.7
HbD	20	36.6 – 43.4	5.8 – 8.6
HbE	20	25.5 – 32.5	5.9 – 8.3
HbA2	25	5.0 - 13.3	5.0 - 14.5
HbF	30	4.1 – 30.2	4.4 - 14.4

Table 20: Hemoglobin Variant Study Bias Results

Hemoglobin Variant	Relative % Bias to Comparative Method	
	Relative %Bias (Range of %Bias) for HbA1c	Relative %Bias (Range of %Bias) for HbA1c
	~6.5%	~9.0%
HbS	-0.6 (-5.8 to 5.5)	-1.5 (-3.3 to -0.1)
HbC	-1.3 (-4.0 to 1.3)	-3.9 (-5.5 to -2.4)
HbD	-4.7 (-6.7 to -1.1)	-4.4 (-6.3 to -.2.4)
HbE	-2.7 (-6.7 to 1.6)	-1.3 (-2.0 to -0.6)
HbA2	-1.3 (-5.1 to 0.5)	3.4 (2.8 to 4.1)
HbF	-2.3 (-4.1 to -0.7)	-3.5 (-4.2 to -2.8)

2. Matrix comparison
The data supports the use of the following blood collection tubes with the D-100™ HbA1c test in Table 21.

Table 21: Anticoagulant

K_2-EDTA
K_3.EDTA
Potassium Oxalate/Sodium Fluoride
Sodium Citrate
Sodium Heparin
Lithium Heparin

3. Expected Values/Reference Range
Hemoglobin A1c expected values range was cited from American Diabetes Association Standards of Medical Care in Diabetes 2010, 33 (Supplement 1), S62-S69 for Diagnosis of Diabetes are presented in Table 22.

Table 22: Hemoglobin A1c Expected Values

Hemoglobin A1c		**Suggested Diagnosis**
NGSP %	**IFCC mmol/mol**	
≥6.5	≥47	Diabetic
5.7 – 6.4	39-46	Pre-Diabetic
<5.7	<39	Non-Diabetic

Conclusion:

The information and data in this 510(k) document demonstrate that the D-100™ HbA1c test as performed on the D-100™Hemoglobin Testing System is an accurate, reliable, precise test that correlates well with current cleared methods and NGSP standardized testing for the quantitation of HbA1c. The contents of this submission demonstrates that the D-100™ HbA1c test as performed on the D-100™ Hemoglobin Testing System is substantially equivalent to its predicate device, VARIANT II TURBO HbA1c Kit – 2.0 and, therefore, safe and effective for its intended use. The performance criteria as stipulated by the Special Controls requirements for HbA1c systems that diagnose diabetes have clearly been met. The D-100™ HbA1c must be found to be substantially equivalent to the predicate and, therefore, cleared by the agency for the intended use requested.