DEPARTMENT OF HEALTH & HUMAN SERVICES Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
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Silver Spring, MD 20993-0002

November 21, 2016

DIAZYME LABORATORIES
ABHIJIT DATTA
DIRECTOR, TECHNICAL OPERATIONS
12889 GREGG COURT
POWAY CA 92064

Re: K160762
Trade/Device Name: Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis),
Diazyme Direct HbA1c Assay Calibrator Set,
Diazyme Direct HbA1c Assay Control Set
Regulation Number: 21 CFR 864.7470
Regulation Name: Glycosylated hemoglobin assay
Regulatory Class: II
Product Code: LCP, JIT, JJX
Dated: October 5, 2016
Received: October 6, 2016

Dear Abhijit Datta:

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)*
k160762

Device Name
Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis), Diazyme Direct HbA1c Assay Calibrator Set, Diazyme Direct HbA1c Assay Control Set

Indications for Use *(Describe)*
Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) test kit is intended for use in the quantitative determination of stable HbA1c in venous whole blood samples with on-board blood lysis application in a clinical laboratory. This test is not to be used to diagnose or screen for diabetes. The measurement of HbA1c concentration is for use in monitoring long-term glucose control of persons with diabetes. For in-vitro diagnostic use only.

Diazyme Direct HbA1c Assay Calibrator Set is intended to be used for calibration of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis). For in-vitro diagnostic use only.

Diazyme Direct HbA1c Assay Control Set is intended to be used for quality control by monitoring accuracy and precision of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis). For in-vitro diagnostic use only.

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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k160762

510k Summary

This summary of 510(k) safety and effectiveness information is being submitted in accordance with the requirements of SMDA 1990 and 21 CFR 807.92.

Submitter's name: Diazyme Laboratories

Submitter's address:
12889 Gregg Court
Poway, CA 92064
USA

Name of Contact Person:
Dr. Abhijit Datta
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12889 Gregg Court
Poway, CA 92064
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Fax: 858-455-2120

Date the Summary was Prepared: November 15, 2016

Name of the Device
Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis)
Diazyme Direct HbA1c Assay Calibrator Set
Diazyme Direct HbA1c Assay Control Set

Trade Name:
Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis)
Diazyme Direct HbA1c Assay Calibrator Set
Diazyme Direct HbA1c Assay Control Set

Common Name: Direct Enzymatic Assay, HbA1c

Classification Name: Glycosylated Hemoglobin Assay

Establishment Registration

The Device Establishment Registration Number is 2032900.

Regulatory Information:

Regulation Description	Product Code	Device Class	Regulation	Panel
Glycosylated Hemoglobin Assay	LCP	II	21 CFR 864.7470	Hematology, 81
Calibrator	JIT	II	21 CFR §862.1150	Chemistry, 75
Quality Control Material	JJX	I	21 CFR §862.1660	Chemistry, 75

Submission type: Traditional 510k

Predicate Device

Diazyme Laboratories claims substantial equivalence to the currently marketed Diazyme Direct HbA1c Enzymatic Assay. The reference numbers are k070743.

Performance Standards

At this time, no special controls under Section 513 or performance standard under Section 514 have been issued for *in vitro* diagnostic products.

Manufacturing Address

The Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) will be manufactured and distributed by Diazyme Laboratories.

Manufacturing Site Address
Diazyme Laboratories
12889 Gregg Court
Poway, CA 92064

Description of the Device

Clinical Significance

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, as part of the diabetes management program, HbA1c testing should be performed at least biannually in all patients and quarterly for patients whose therapy has changed or who are not meeting treatment goals. 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.

Assay Principle

Diazyme Direct HbA1c test is an enzymatic assay in which lysed whole blood samples are subjected to extensive protease digestion with Bacillus sp protease. This process releases amino acids including glycated valines from the hemoglobin beta chains. Glycated valines then serve as substrates for specific recombinant fructosyl valine oxidase (FVO) enzyme. The recombinant FVO specifically cleaves N-terminal valines and produces hydrogen peroxide. This, in turn, is measured using a horseradish peroxidase (POD) catalyzed reaction and a suitable chromogen. No separate measurement for total Hemoglobin (Hb) is needed in this Direct HbA1c Assay.

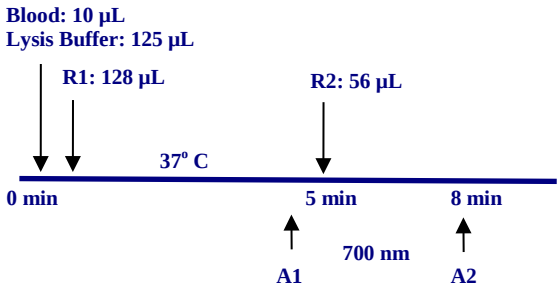
Detailer Device Description

1. The Diazyme Direct HbA1c assay kit (Enzymatic, On-Board Lysis) is comprised of a Reagent 1, Reagent 2, Lysis Buffer.
2. Diazyme Direct HbA1c Assay Calibrator Set: 2 levels, human whole blood based in lyophilized form. It is intended to be used for calibration of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis).
3. Diazyme Direct HbA1c Assay Control Set: 2 levels, human whole blood based in lyophilized form. It is intended to be used for quality control by monitoring accuracy and precision of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis).

The verification and/or validation studies were performed on the automated chemistry analyzer Roche Modular P (k953239/005). Modular P analyzer is a high through-put automated chemistry analyzer and that can process 800 samples per hour. The predicate performance studies at submission were performed on Hitachi 917 which is an obsolete member of the Roche Mod P Roche family of instruments. The Control Unit of the Roche Modular P analyzer uses a graphical interface to control all instrument functions. The computer, keyboard, and touchscreen monitor allows users to navigate through the software, enter assay, calibrator, and control information, and make test selections. The Diazyme Direct HbA1c Assay Modular P application parameters provided are programmed into the Modular P analyzers. The reagents, calibrators and controls are loaded into the analyzer. The Roche Modular P stores the Diazyme Direct HbA1c Assay reagents in a refrigerated compartment. Reagent and sample pipettes automatically aspirate and dispense specified amounts of reagents or calibrators, controls, and samples into reaction cells. The change in absorbance is measured at specified wavelengths.

Assay Scheme for Roche Modular P Analyzer

Use the following scheme as a guideline for analyzer application. Note: Diazyme Direct HbA1c Assay is an end-point assay and the first reading point A1 is right before the addition of reagent R2.



After a 2-point calibration, linear-fitting is used to fit calibration curve through mean values of the response for calibrators of known concentrations. The Roche Modular P calculates the HbA1c% concentration of a patient sample by interpolation of the obtained signal to a stored 2-point calibration curve.

#: The Modular P analyzer performs automatic on-board 1: 12.5 dilution using lysis buffer (10 µL sample + 125 µL lysis buffer) for rapid blood sample lysis.

The following key parameters are input into the Mod P user defined channel interface using the provided Diazyme Direct HbA1c Assay (DZ168C) Parameter sheet.

Modular P Applications
Measuring mode: Absorbance
Abs. calculation mode: 2 Point END
Reaction mode: S-R1/R3
Reaction direction: Increase
Primary/Secondary Wavelength: 700nm/800nm
Calc. first/last: 16/28
Unit: %
Pipetting parameters
R1: 128 µL
Sample: 10 µL; Sample Diluent (lysis buffer) 125 µL (1:12.5 dilution)
Diluted Sample: 20 µL
R2: 56 µL
Total volume: 204 µL

The Design control verification and/or validation studies were performed on Roche Mod P automated analyzer.

Intended Use of the Device:
Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) test kit is intended for use in the quantitative determination of stable HbA1c in venous whole blood samples with on-board blood lysis application in a clinical laboratory. This test is not to be used to diagnose or screen for

diabetes. The measurement of HbA1c concentration is for use in monitoring long-term glucose control of persons with diabetes. For in-vitro diagnostic use only.

Diazyme Direct HbA1c Assay Calibrator Set is intended to be used for calibration of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis). For in-vitro diagnostic use only.

Diazyme Direct HbA1c Assay Control Set is intended to be used for quality control by monitoring accuracy and precision of Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis). For in-vitro diagnostic use only.

Performance Characteristics of Device

Precision Summary

1) Internal Precision

Internal precision of the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) was evaluated according to CLSI EP5-A2 guideline. This assessment was performed on the Roche Modular P analyzer at Diazyme Laboratories.

In the study, three lots of reagents were used. For each lot of reagent, 5 unaltered whole blood specimens containing 4.6%, 5.4%, 7.5%, 9.7% and 11.9% HbA1c spanning AMR were tested. Three lots of HbA1c calibrators and three lots of HbA1c controls, two levels each, were tested as samples to evaluate precision. All samples were tested 2 runs per day, 2 replicates per run over 20 working days according to CLSI EP5-A2 protocol. Whole blood samples used in the study were IRB approved.

Precision data for the three lots of reagents tested are given below. Data analyzed with EP Evaluator (Release 11).

Sample	Mean (N=240)	Within-Run		Between-Run		Between-day		Between lot		Total	
		SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Sample 1	4.64	0.04	0.8%	0.07	1.5%	0.00	0.0%	0.08	1.6%	0.08	1.7%
Sample 2	5.36	0.05	0.9%	0.05	0.9%	0.00	0.0%	0.07	1.2%	0.07	1.2%
Sample 3	7.51	0.05	0.6%	0.05	0.7%	0.00	0.0%	0.07	0.9%	0.07	0.9%
Sample 4	9.61	0.06	0.6%	0.05	0.5%	0.03	0.3%	0.08	0.9%	0.08	0.9%
Sample 5	11.89	0.09	0.7%	0.08	0.6%	0.04	0.4%	0.12	1.0%	0.12	1.0%
Con 1 Lot 1	6.22	0.05	0.8%	0.03	0.5%	0.01	0.1%	0.06	1.0%	0.06	1.0%
Con 2 Lot 1	9.47	0.06	0.6%	0.04	0.4%	0.02	0.2%	0.07	0.8%	0.07	0.8%
Con 1 Lot 2	5.70	0.04	0.8%	0.04	0.7%	0.02	0.4%	0.06	1.1%	0.06	1.1%
Con 2 Lot 2	9.11	0.05	0.5%	0.04	0.4%	0.03	0.3%	0.07	0.7%	0.07	0.7%
Con 1 Lot 3	6.04	0.05	0.7%	0.05	0.9%	0.00	0.0%	0.07	1.1%	0.07	1.2%
Con 2 Lot 3	9.68	0.05	0.6%	0.04	0.4%	0.00	0.0%	0.07	0.7%	0.07	0.7%
Cal 1 Lot 1	6.22	0.04	0.6%	0.04	0.6%	0.00	0.0%	0.05	0.8%	0.05	0.9%
Cal 2 Lot 1	12.30	0.07	0.6%	0.04	0.3%	0.03	0.2%	0.09	0.7%	0.09	0.7%
Cal 1 Lot 2	5.68	0.05	0.9%	0.04	0.7%	0.02	0.4%	0.07	1.1%	0.07	1.2%
Cal 2 Lot 2	9.70	0.06	0.6%	0.04	0.4%	0.02	0.2%	0.07	0.8%	0.07	0.8%

Cal 1 Lot 3	6.04	0.05	0.8%	0.04	0.7%	0.00	0.0%	0.06	1.0%	0.06	1.1%
Cal 2 Lot 3	11.30	0.06	0.5%	0.04	0.3%	0.01	0.1%	0.07	0.6%	0.07	0.6%

Conclusion: 20-day reproducibility data for all samples tested showed that the within-run, between-run, between-day, between-lot and total CV% were less than 2%, meeting acceptance criteria.

2) Multiple Site Precision Study (Three sites)

Multiple site precision study was performed at two external sites and the Diazyme site. In this study, same sets of samples were used at all sites. Multiple site precision of the Diazyme Direct HbA1c Assay was evaluated according to a modified CLSI EP5-A2 guideline on Roche Modular P analyzer. Five whole blood samples containing 4.6%, 5.4%, 7.5%, 9.7% and 11.9% HbA1c spanning assay AMR, three lots of calibrators and three lots of controls were tested in duplicates per run, 2 runs per day for 5 working days with the same lot of the reagent to evaluate between site assay precision. Testing was conducted at 3 different sites, by 3 different operators, on 3 different Modular P instruments. Within-run, between-run, between-day, between-site, and total precision were calculated.

Multiple site precision data is summarized in the following table:

Sample	Mean (n=60)	Within-Run		Between-Run		Between-day		Total	
		SD	%CV	SD	%CV	SD	%CV	SD	%CV
Sample 1	4.67	0.05	1.0%	0.04	0.8%	0.02	0.5%	0.07	1.4%
Sample 2	5.37	0.04	0.8%	0.05	1.0%	0.00	0.0%	0.07	1.2%
Sample 3	7.52	0.05	0.7%	0.06	0.8%	0.00	0.0%	0.08	1.0%
Sample 4	9.67	0.07	0.8%	0.11	1.1%	0.00	0.0%	0.13	1.4%
Sample 5	11.92	0.09	0.8%	0.09	0.8%	0.06	0.5%	0.14	1.2%
Con1 Lot 1	6.21	0.03	0.6%	0.00	0.0%	0.00	0.0%	0.03	0.6%
Con2 Lot 1	9.48	0.05	0.6%	0.05	0.6%	0.03	0.3%	0.08	0.9%
Con1 Lot 2	5.63	0.04	0.7%	0.02	0.4%	0.03	0.6%	0.06	1.0%
Con2 Lot 2	9.11	0.05	0.5%	0.04	0.4%	0.03	0.3%	0.07	0.7%
Con1 Lot 3	6.01	0.05	0.8%	0.01	0.2%	0.02	0.3%	0.06	0.9%
Con2 Lot 3	9.65	0.06	0.7%	0.05	0.5%	0.04	0.4%	0.09	1.0%
Cal1 Lot 1	6.21	0.04	0.7%	0.03	0.5%	0.00	0.0%	0.06	0.9%
Cal2 Lot 1	12.32	0.07	0.6%	0.05	0.4%	0.05	0.4%	0.10	0.8%
Cal1 Lot 2	5.63	0.06	1.0%	0.00	0.0%	0.02	0.3%	0.06	1.1%
Cal2 Lot 2	9.74	0.06	0.6%	0.06	0.7%	0.03	0.4%	0.10	1.0%
Cal1 Lot 3	6.02	0.05	0.8%	0.03	0.4%	0.01	0.1%	0.05	0.9%
Cal2 Lot 3	11.36	0.06	0.6%	0.08	0.7%	0.05	0.5%	0.12	1.0%

Sample	Mean	Between-Site	
	(n=60)	SD	%CV
Sample 1	4.7	0.07	1.4
Sample 2	5.4	0.06	1.2
Sample 3	7.5	0.07	0.9
Sample 4	9.7	0.12	1.3

Sample 5	11.9	0.14	1.2
Control 1 Lot 1	6.2	0.03	0.5
Control 2 Lot 1	9.5	0.08	0.9
Control 1 Lot 2	5.6	0.06	1.0
Control 2 Lot 2	9.1	0.07	0.7
Control 1 Lot 3	6.0	0.06	0.9
Control 2 Lot 3	9.6	0.09	0.9
Calibrator 1 Lot 1	6.2	0.05	0.8
Calibrator 2 Lot 1	12.3	0.10	0.8
Calibrator 1 Lot 2	5.6	0.05	1.0
Calibrator 2 Lot 2	9.7	0.10	1.0
Calibrator 1 Lot 3	6.0	0.05	0.9
Calibrator 2 Lot 3	11.4	0.12	1.0

Conclusion: All samples tested showed that the within-run precision, between-run, between-day, between-site, and total CV% were less than 2% meeting the acceptance criteria.

LOD/LOB/LOQ

The LOB, LOD, and LOQ of the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis Assay were determined according to CLSI EP17-A2 with three lots of the reagents.

The LOB was determined as 0.0%, 0.1% and 0.2% for reagent lot 1, lot 2, and lot 3 respectively. The HbA1c Assay LOB is obtained as the maximal value between all three reagents lots, 0.2%.

The LOD was determined as 0.38%, 0.43%, and 0.5% for reagent lot 1, lot 2, and lot 3 respectively. The Direct HbA1c Assay LOD is obtained as the maximal value among the three lots, 0.5%.

Using the calculated CV's from the 5 concentrations, curve fit analysis was performed to determine the upper 95% confidence level of point with CV less than 20%. EP Evaluator (Release 11) analysis showed LOQ = 0.8% for Reagent Lots 1, 2, and 3, respectively.

Linearity

11 levels of whole blood linearity set were prepared by dilution of two whole blood samples, Low and high samples with known HbA1c of 3.8% and 12.3% respectively were used to generate the test levels according to CLSI EP6-A and tested in triplicate, again, per guideline. Data analysis showed bias ≤10%. The assay is linear from 4% to12% HbA1c.

Calibrator Traceability

Diazyme Direct HbA1c Assay is traceable to NGSP/DCCT. The assay methodology is traceable to IFCC per NGSP correlation guideline (www.ngsp.org).

Method Comparison Study

Following the CLSI: EP9-A2 guideline, the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) was evaluated on the Roche Modular P testing individual K2 EDTA whole blood samples (IRB approved) in comparison with Tosoh G8 HPLC HbA1c method, which is used as the NGSP certification method in NGSP reference laboratories (www.NGSP.org). A total of 124 unique samples at site 1, total of 132 unique samples at site 2, and a total of 120 unique samples at site 3 were tested using the same lot of the reagents at three different sites on three different Modular P analyzers by three different operators.

Summary of Regression Analysis: Regular Linear

Parameter	Site 1	Site 2	Site 3	Combined
Slope	1.006	1.033	1.026	1.023
95% CI	0.983 – 1.030	1.015 – 1.051	1.005 – 1.047	1.011 – 1.035
Intercept	0.04	-0.126	-0.18	-0.090
95% CI	-0.13 to 0.21	-0.268 to 0.015	-0.34 to -0.01	-0.180 to 0.000
Correlation Coefficient(R)	0.9918	0.9950	0.9938	0.9937
Sample Range (Diazyme)	4.4 – 12.0	4.2 - 11.9	4.4 – 12.0	4.2 – 12.0

Summary of Regression Analysis: Deming

Parameter	Site 1	Site 2	Site 3	Combined
Slope	1.015	1.039	1.033	1.030
95% CI	0.992 – 1.038	1.021 – 1.057	1.012 – 1.054	1.018 – 1.042
Intercept	-0.02	-0.167	-0.23	-0.140
95% CI	-0.19 to 0.15	-0.309 to -0.026	-0.40 to -0.06	-0.230 to -0.050
Correlation Coefficient(R)	0.9918	0.9950	0.9938	0.9937
Sample Range (Diazyme)	4.4 – 12.0	4.2 - 11.9	4.4 – 12.0	4.2 – 12.0

Conclusion: Method comparison data analysis of the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) versus the predicate Tosoh HPLC Assay testing K2 EDTA whole blood at three different sites on three different Modular P analyzers by three different operators demonstrated the results met the acceptance criteria.

Analytical specificity

Endogenous substances:

To determine the level of interference from substances present whole blood samples, the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) was used to test three whole blood samples, which contained "low", "medium", and "high" HbA1c concentrations, following CLSI EP7-A2. To evaluate interference, each whole blood samples were spiked with potential endogenous interference substances per protocol and tested in triplicates on the Roche Modular P analyzer.

The following substances showed no significant interference (≤ ±10% deviation) up to the concentrations summarized below.

Interfering Substances	Concentration
Ascorbic Acid	12 mg/dL
Bilirubin	15 mg/dL
Bilirubin Conjugated	13 mg/dL
Hemoglobin	21 mg/dL
Triglycerides	4000 mg/dL
Glucose	4000 mg/dL
Uric Acid	30 mg/dL
Urea	80 mg/dL
Acetaminophen	20 mg/dL
Acetylsalicylic Acid	65.2 mg/dL
Metformin	4 mg/dL
Ibuprofen	50 mg/dL
Glyburide	0.19 mg/dL
Total Protein	21000 mg/dL
Vitamin E	13.6 mg/dL
Rheumatoid Factor	375 IU/mL

Additionally acetylated, carbamylated and labile HbA1c does not adversely affect the assay. Hemoglobinopathies may interfere with glycated hemoglobin analysis. Testing results indicate that there is no significant interference for Hemoglobin C (≤ 38.2%), Hemoglobin D (≤ 43.1%), Hemoglobin E (≤ 21.1%), and Hemoglobin S (≤ 37.3%). High HbF (>10%) may result in inaccurate HbA1c values.

Substantial Equivalence

Device Modifications

The intended use of the modified device and fundamental scientific technology used in the test has not changed as a result of the modification.

Specific modifications:

1. Change in number of reagents supplied with on-board lysis application
2. Change in concentration of reagent components
3. Change in product labeling to reflect the modification

Summary of Assay Kit Components

Predicate device k070734	Subject Device
Lysis buffer	Lysis buffer
100mM CHES pH 8.7, 1% Triton X 100, 0.45% SDS, 0.5mM redox agent	>100mM Tris pH >8.0, 1% Triton X 100, >1.5% nonionic and ionic detergents, >4KU/mL proteases
Reagent1a	Reagent1
5mM MES pH 7.0, >4KU/mL proteases, >10μM redox agent	5mM MES pH >6.0, <3mM redox agent
Reagent 1b	N/A
1mM MES pH 6.3, <3mM redox agent	
Reagent 2	Reagent 2
15mM Tris pH8.0, >10U/mL FVO enzyme, 90U/mL POD, 0.8mM chromagen	>5mM bis-Tris pH>7.0, >10U/mL FVO enzyme, 90U/mL POD, >50μM chromagen
None	None
HbA1c Calibrator set (linear mode, lyophilized whole blood based)	HbA1c Calibrator set (linear mode, lyophilized whole blood based)
1 x 0.5mL Level 1	1 x 0.5mL Level 1
1 x 0.5mL Level 2	1 x 0.5mL Level 2
HbA1c Control set (lyophilized whole blood based)	HbA1c Control set (lyophilized whole blood based)
1 x 0.5mL Level 1	1 x 0.5mL Level 1
1 x 0.5mL Level 2	1 x 0.5mL Level 2

Substantial Equivalence Comparison Table

Indications for Use

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis) (modified device; special 510k submission)	Equivalency
Diazyme Direct Enzymatic Hemoglobin	Diazyme Direct HbA1c Assay	Same

A1c (glycated hemoglobin A1c; A1c; HbA1c) reagents are intended for use in the quantitative determination of stable HbA1c in human whole blood samples. Measurement of hemoglobin A1c is a valuable indicator for long-term diabetic control. For *in-vitro* diagnostic use only.	(Enzymatic, On-Board Lysis) test kit is intended for use in the quantitative determination of stable HbA1c in venous whole blood samples with on-board blood lysis application in a clinical laboratory. This test is not to be used to diagnose or screen for diabetes. The measurement of HbA1c concentration is for use in monitoring long-term glucose control of persons with diabetes. For in-vitro diagnostic use only.	

Principle

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
Direct Enzymatic HbA1c test is an enzymatic assay in which lysed whole blood samples are subjected to extensive protease digestion with Bacillus *sp* protease. This process releases amino acids including glycated valines from the hemoglobin beta chains. Glycated valines then serve as substrates for specific recombinant fructosyl valine oxidase (FVO) enzyme, produced in *E. coli*. The recombinant FVO specifically cleaves N-terminal valines and produces hydrogen peroxide. This, in turn, is measured using a horse radish peroxidase (POD) catalyzed reaction and a suitable chromagen. The HbA1c concentration is expressed directly as %HbA1c by use of a suitable calibration curve in which the calibrators have values for each level in %HbA1c.	Diazyme Direct HbA1c Assay is an enzymatic assay in which lysed whole blood samples are subjected to extensive protease digestion with Bacillus sp protease. This process releases amino acids including glycated valines from the hemoglobin beta chains. Glycated valines then serve as substrates for specific recombinant fructosyl valine oxidase (FVO) enzyme. The recombinant FVO specifically cleaves N-terminal valines and produces hydrogen peroxide. This, in turn, is measured using a horseradish peroxidase (POD) catalyzed reaction and a suitable chromogen. The HbA1c concentration is expressed directly as %HbA1c by use of a suitable calibration curve in which the calibrators have values for each level in %HbA1c.	Same

Test Objective

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
For the *in vitro* quantitative determination of % HbA1c in human whole blood.	For the *in vitro* quantitative determination of % HbA1c in human whole blood.	Same

Type of Test

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
Quantitative	Quantitative	Same

Specimen Type

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
EDTA Human whole blood.	K2 EDTA Human whole blood.	Same

Specimen Lysis

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
Human whole blood Specimens are lysed manually off-board	Human whole blood specimens are lysed on-board on analyzer	Different

Product Type

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
3 Reagents plus lysis buffer, Calibrators, Controls	2 Reagents plus lysis buffer, Calibrators, Controls	Different

Performance Comparison

Direct HbA1c Enzymatic Assay (k070734)	Direct HbA1c Assay (Enzymatic, On-Board Lysis)	Equivalency
Linear Range: 4-16% HbA1c[1] Total Precision: 1.8% Method Comparison: Correlation Coefficient: 0.9874 Slope/Intercept: y = 1.0212x + 0.0135	Linear Range: 4-12 % HbA1c[1] Total Precision: 0.6-1.9% Method Comparison: Correlation Coefficient: 0.9937 Slope/Intercept: y = 1.023x - 0.09	Substantially similar

[1]Linear Range limited to 4-12% HbA1c so that the upper limit of AMR is not too far from NGSP protocol for certification sample range.

Summary of Rationale for Considering the Device Substantially Equivalent to Devices Approved for Interstate Commerce

In summary, the Intended Use of the modified device has not changed. The assay principle and fundamental scientific technology of the modified device has not changed. The major changes are in the number of reagents supplied (from 3 reagent plus lysis buffer to 2 reagents plus lysis buffer), Concentration of components in the reagents and labeling changes reflecting the modification. Design control activities including precision, limit of quantitation, linearity, accuracy and interference studies demonstrates that the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) is substantially similar to the predicate device (k070734) and is safe and effective. There is no significant deviation between performance of the modified device Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) and the predicate device. The differences in the assay steps should not affect the safety and effectiveness of the Diazyme Direct Hba1c assay (Enzymatic, On-Board Lysis). Diazyme Direct HbA1c assay calibrator is traceable to NGSP certification for the modified device. The legally marketed device Tosoh HPLC HbA1c Test is used as standard NGSP certification method in NGSP reference laboratories and was thus selected for accuracy study. The Tosoh HPLC HbA1c Test is also the device used in accuracy studies for the predicate Diazyme HbA1c Enzymatic assay device (k070734). The accuracy studies with whole blood samples tested with the modified device unequivocally demonstrated excellent correlation. In conclusion, the Diazyme Direct HbA1c Assay (Enzymatic, On-Board Lysis) for measurement of HbA1c in patient whole blood samples is substantially equivalent to legally marketed devices. The differences in the assay steps should not affect the safety and effectiveness of the Diazyme Direct HbA1c assay (Enzymatic, On-Board Lysis) and risk analysis performed indicates no change in risk stance. The Diazyme Direct HbA1c assay gives similar diagnostic information as the other legally marketed HbA1c tests and offers users a rapid and convenient device for measuring HbA1c in human blood.