March 26, 2020

Roche Diagnostics Operations Inc.
Leslie Patterson
Regulatory Affairs Principal
9115 Hague Rd
Indianapolis, Indiana 46250

Re: K193053
Trade/Device Name: Tina-quant Hemoglobin A1cDx Gen.3
Regulation Number: 21 CFR 862.1373
Regulation Name: Hemoglobin A1c Test System
Regulatory Class: Class II
Product Code: PDJ, LCP
Dated: February 13, 2020
Received: February 14, 2020

Dear Leslie Patterson:

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. Although this letter refers to your product as a device, please be aware that some cleared products may instead be combination products. The 510(k) Premarket Notification Database located at https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpmn/pmn.cfm identifies combination product submissions. 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 Part 801 and Part 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR

803) for devices or postmarketing safety reporting (21 CFR 4, Subpart B) for combination products (see https://www.fda.gov/combination-products/guidance-regulatory-information/postmarketing-safety-reporting-combination-products); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820) for devices or current good manufacturing practices (21 CFR 4, Subpart A) for combination products; and, if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

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 https://www.fda.gov/medical-devices/medical-device-safety/medical-device-reporting-mdr-how-report-medical-device-problems.

For comprehensive regulatory information about medical devices and radiation-emitting products, including information about labeling regulations, please see Device Advice (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance) and CDRH Learn (https://www.fda.gov/training-and-continuing-education/cdrh-learn). Additionally, you may contact the Division of Industry and Consumer Education (DICE) to ask a question about a specific regulatory topic. See the DICE website (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance/contact-us-division-industry-and-consumer-education-dice) for more information or contact DICE by email (DICE@fda.hhs.gov) or phone (1-800-638-2041 or 301-796-7100).

Sincerely,

Marianela Perez-Torres, Ph.D.
Acting Deputy Director
Division of Chemistry and Toxicology Devices
OHT7: Office of In Vitro Diagnostics and Radiological Health
Office of Product Evaluation and Quality
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: 06/30/2020
See PRA Statement below.

510(k) Number *(if known)*
k193053

Device Name
Tina-quant Hemoglobin A1cDx Gen.3

Indications for Use *(Describe)*
The Tina-quant Hemoglobin A1cDx Gen.3 assay is intended for use as an aid in diagnosis of diabetes and as an aid in identifying patients who may be at risk for developing diabetes. It is an in vitro diagnostics reagent system intended for quantitative determination of mmol/mol hemoglobin A1c (IFCC) and % hemoglobin A1c (DCCT/NGSP) in hemolysate or venous whole blood on the cobas c 503 clinical chemistry analyzer. HbA1c determinations are useful for monitoring of long-term blood glucose control in individuals with diabetes mellitus.

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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Tina-quant Hemoglobin A1cDx Gen.3
510(k) Summary (k193053)

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

In accordance with 21 CFR 807.87, Roche Diagnostics hereby submits official notification as required by Section 510(k) of the Federal Food, Drug and Cosmetics Act of our intention to market the device described in this Premarket Notification 510(k).

The purpose of this Traditional 510(k) Premarket Notification is to obtain FDA review and clearance for the Tina-quant Hemoglobin A1cDx Gen.3 assay.

Submitter Name	Roche Diagnostics Operations Inc.
Address	9115 Hague Road P.O. Box 50416 Indianapolis, IN 46250-0457
Contact	Leslie Patterson Phone: (317) 521-7307 FAX: (317) 521-2324 Email: leslie.patterson@roche.com
Date Prepared	February 13, 2020
Proprietary Name	Tina-quant Hemoglobin A1cDx Gen.3
Common Name	Glycosylated Hemoglobin Assay
Classification Name	Hemoglobin A1c test system
Product Codes, **Regulation Numbers**	PDJ, 862.1373 LCP, 864.7470
Predicate Device	COBAS INTEGRA 800 Tina-quant Hemoglobin A1cDx Gen.2 assay, k121291
Establishment Registration	1823260, Roche Diagnostics Corporation

1. DEVICE DESCRIPTION

Tina-quant Hemoglobin A1cDx Gen.3 assay is an in vitro diagnostics reagent system intended for quantitative determination of mmol/mol hemoglobin A1c (IFCC) and % hemoglobin A1c (DCCT/NGSP) in hemolysate or whole blood on the **cobas c** 503 clinical chemistry analyzer.

The assay offers separate applications that are specific to the sample types whole blood and hemolysate. The Whole Blood Application differs from the Hemolysate Application in the hemolyzing step. For the Whole Blood Application, whole blood samples are placed on the analyzer and hemolysis occurs onboard the analyzer. For the Hemolysate Application, hemolyzed samples are placed on the analyzer and hemolysis occurs manually before placing the samples onboard the analyzer. The two applications yield the same results. Hemolyzing reagent is part of the test system and is either placed on board the analyzer for the Whole Blood Application or used manually for the Hemolysate Application.

Anticoagulated whole blood is hemolyzed either manually or automatically prior to determination of HbAlc by a turbidimetric inhibition immunoassay. Liberated hemoglobin (Hb) in the hemolyzed sample is converted to a derivative having a characteristic absorption spectrum and measured bichromatically. The instrument calculates the % HbAlc from the HbAlc/Hb ratio according to a user selected protocol, either IFCC or NGSP protocols.

1.1. Test Principle

This method uses tetradecyltrimethylammonium bromide (TTAB) as the detergent in the hemolyzing reagent to eliminate interference from leukocytes (TTAB does not lyse leukocytes). Sample pretreatment to remove labile HbA1c is not necessary. All hemoglobin variants which are glycated at the β -chain N-terminus and which have antibody-recognizable regions identical to that of HbA1c are determined by this assay. Consequently, the metabolic state of patients having uremia or the most frequent hemoglobinopathies (HbAS, HbAC, HbAE, HbAD) can be determined using this assay.

1.2. Hemoglobin A1c

The HbA1c determination is based on the turbidimetric inhibition immunoassay (TINIA) for hemolyzed whole blood.

- Sample and addition of R1 (buffer/antibody):

Glycohemoglobin (HbA1c) in the sample reacts with anti-HbA1c antibody to form soluble antigen-antibody complexes. Since the specific HbA1c antibody site is present only once on the HbA1c molecule, formation of insoluble complexes does not take place.

- Addition of R3 (buffer/polyhapten) and start of reaction:

The polyhaptens react with excess anti-HbA1c antibodies to form an insoluble antibody-polyhapten complex which can be determined turbidimetrically.

1.3. Hemoglobin

Liberated hemoglobin in the hemolyzed sample is converted to a derivative having a characteristic absorption spectrum which is measured bichromatically during the preincubation phase (sample + R1) of the above immunological reaction. A separate Hb reagent is consequently, not necessary.

1.4. Final HbA1c Result

The final result is expressed as mmol/mol HbA1c or % HbA1c and is calculated from the HbA1c/Hb ratio as follows:

Protocol 1 (mmol/mol HbA1c acc. to IFCC): HbA1c (mmol/mol) = (HbA1c/Hb) × 1000

Protocol 2 (% HbA1c acc. to DCCT/NGSP): HbA1c (%) = (HbA1c/Hb) × 91.5 + 2.15

1.5. Standardization

Traceability: This method has been standardized against the approved IFCC reference method for the measurement of HbA1c in human blood and can be transferred to results traceable to DCCT/NGSP by calculation.

2. INDICATIONS FOR USE

The Tina-quant Hemoglobin A1cDx Gen.3 assay is intended for use as an aid in diagnosis of diabetes and as an aid in identifying patients who may be at risk for developing diabetes. It is an in vitro diagnostics reagent system intended for quantitative determination of mmol/mol hemoglobin A1c (IFCC) and % hemoglobin A1c (DCCT/NGSP) in hemolysate or venous whole blood on the **cobas c** 503 clinical chemistry analyzer. HbA1c determinations are useful for monitoring of long-term blood glucose control in individuals with diabetes mellitus.

3. TECHNOLOGICAL CHARACTERISTICS

The following table compares the Tina-quant Hemoglobin A1cDx Gen.3 assay with its predicate device, COBAS INTEGRA 800 Tina-quant Hemoglobin A1cDx Gen.2 (k121291).

Table 1: Substantial Equivalence Assay Comparison

Feature	Predicate Device: COBAS INTEGRA 800 Tina-quant Hemoglobin A1cDx Gen.2 k121291	Submitted Device: Tina-quant Hemoglobin A1cDx Gen.3
Intended Use	This test is to be used as an aid in diagnosis of diabetes and as an aid in identifying patients who may be at risk for developing diabetes. It is an in vitro diagnostic reagent system intended for quantitative determination of mmol/mol hemoglobin A1c (IFCC) and % hemoglobin A1c (DCCT/NGSP) in hemolysate or venous whole blood. HbA1c determinations are useful for monitoring of long-term blood glucose control in individuals with diabetes mellitus.	Same
Sample Types	Anticoagulated venous blood Acceptable anticoagulants for both the hemolysate and whole blood applications include: • Li-Heparin • K_2-EDTA • K_3-EDTA • Fluoride/potassium oxalate • Na-Heparin • NaF/Na_2-EDTA	Anticoagulated venous blood Acceptable anticoagulants for both the hemolysate and whole blood applications include: • Li-Heparin • K_2-EDTA • K_3-EDTA • Fluoride/potassium oxalate • Na-Heparin • EDTA Fluoride
Instrument Platform	COBAS Integra 800 • Absorbance Photometry	**cobas c** 503 Same
Calibrator	Cfas HbA1c	Same

Feature	Predicate Device: COBAS INTEGRA 800 Tina-quant Hemoglobin A1cDx Gen.2 k121291	Submitted Device: Tina-quant Hemoglobin A1cDx Gen.3
Calibration Frequency	• After 29 days on-board the analyzer • After reagent lot change • As required following quality control procedures	Same
Calibration Mode	Logit/Log 5	Hb: linear HbA1c : RCM4
Controls	PreciControl HbA1c norm PreciControl HbA1c path	Same
Reagent Stability	Unopened: • 2-8 ºC until expiration date On-board in use: • 2-8 ºC for 28 days	Same
Reporting Units	• % HbA1c (NGSP/DCCT) • mmol/mol (IFCC)	Same
Antibody	Polyclonal anti-HbA1c from sheep blood	Same
Test Principle	The anticoagulated whole blood specimen is hemolyzed automatically on the COBAS INTEGRA 800 analyzers with COBAS INTEGRA Hemolyzing Reagent Gen.2. This method uses TTAB as the detergent in the hemolyzing reagent to eliminate interference from leukocytes (TTAB does not lyse leukocytes). Sample pretreatment to remove labile HbA1c is not necessary. All hemoglobin variants which are glycated at the β-chain N-terminus and which have antibody-recognizable regions identical to that of HbA1c are determined by this assay. Consequently, the metabolic state of diabetic patients having uremia or the most frequent hemoglobinopathies (HbAS, HbAC, HbAE, HbAD) can be determined by this assay.	This method uses tetradecyltrimethylammonium bromide (TTAB) as the detergent in the hemolyzing reagent to eliminate interference from leukocytes (TTAB does not lyse leukocytes). Sample pretreatment to remove labile HbA1c is not necessary. All hemoglobin variants which are glycated at the β -chain N-terminus and which have antibody-recognizable regions identical to that of HbA1c are determined by this assay. Consequently, the metabolic state of patients having uremia or the most frequent hemoglobinopathies (HbAS, HbAC, HbAE, HbAD) can be determined using this assay.
Determination of HbA1c	Turbidimetric immunoinhibition (TINIA). Antigen-antibody complexes are formed and excess Ab aggregate with polyhapten to form insoluble complexes	HbA1c determination is based on the turbidimetric inhibition immunoassay (TINIA) for hemolyzed whole blood. Glycohemoglobin in the sample reacts with anti-HbA1c antibody to form soluble antigen-antibody complexes. Polyhaptens react with excess anti-HbA1c antibodies to form an insoluble antibody-polyhapten complex which can be measured turbidimetrically.
Determination of Hb	Bichromatic photometric determination after conversion to a colored derivate	Liberated hemoglobin in the hemolyzed sample is converted to a derivative having a characteristic absorption spectrum which is measured bichromatically.

Feature	Predicate Device: COBAS INTEGRA 800 Tina-quant Hemoglobin A1cDx Gen.2 k121291	Submitted Device: Tina-quant Hemoglobin A1cDx Gen.3
Determination of % HbA1c	The final result is expressed as % HbA1c and is calculated from the HbA1c/Hb ratio per DCCT/NGSP as follows: HbA1c (%) = (HbA1c/Hb) × 91.5 + 2.15	Same
Measuring Range	Hemoglobin: 4-35 g/dL HbA1c: 0.3-3.4 g/dL This corresponds to a measuring range of 23-258 mmol/mol HbA1c (IFCC) and 4.3-24.8 % HbA1c (DCCT/NGSP) at a typical hemoglobin concentration of 13.2 g/dL.	Hemoglobin: 4-40 g/dL (2.48-24.8 mmol/L) HbA1c: 0.3-2.6 g/dL (0.186-1.61 mmol/L) This corresponds to a measuring range of 23-196 mmol/molHbA1c (IFCC) and 4.2-20.1 % HbA1c (DCCT/NGSP) at a typical hemoglobin concentration of 13.2 g/dL (8.2 mmol/L).
Traceability	The assigned HbA1c and total hemoglobin values of the **cobas c** Tina-quant Hemoglobin A1cDx Gen.3 assay is certified with the National Glycohemoglobin Standardization Program (NGSP). NGSP certification is repeated annually.	Same
Reagent Composition	R1 Antibody Reagent: • MES (2-morpholinoethane sulfonic acid) buffer: 0.025 mol/L • TRIS (Tris(hydroxymethyl) aminomethane) buffer: 0.015 mol/L, pH 6.2 • HbA1c antibody (ovine serum): ≥ 0.5 mg/ml • detergents; stabilizers; preservatives SR Polyhapten Reagent: • MES buffer: 0.025 mol/L • TRIS buffer: 0.015 mol/L, pH 6.2 • HbA1c polyhapten: > 8µg/mL • stabilizers; preservatives A1CD (Hemolyzing Reagent): • Aqueous buffered matrix, pH 7.25 • Tetradecyltrimethylammonium bromide: 36 g/L • sodium dihydrogenphosphate monohydrate: 16 mmol/L • sodium monohydrogenphosphate dihydrate: 64 mmol/L stabilizers; preservatives	R1 Antibody Reagent: • MES (2-morpholinoethane sulfonic acid) buffer: 0.025 mol/L • TRIS (Tris(hydroxymethyl)aminomethane) buffer: 0.015 mol/L, pH 6.2 • HbA1c antibody (ovine serum): ≥ 0.5 mg/ml • detergents; stabilizers; preservatives R3 Polyhapten Reagent: • MES buffer: 0.025 mol/L • TRIS buffer: 0.015 mol/L, pH 6.2 • HbA1c polyhapten: > 8 µg/mL • detergents; stabilizers; preservatives A1CD (Hemolyzing Reagent): • Aqueous buffered matrix, pH 7.25 • Tetradecyltrimethylammonium bromide: 36 g/L • Sodium dihydrogenphosphate monohydrate: 16 mmol/L • Sodium monohydrogenphosphate dihydrate: 64 mmol/L stabilizers; preservatives

4. NON-CLINICAL PERFORMANCE EVALUATION

Performance characteristics were evaluated with Tina-quant Hemoglobin A1cDx Gen.3 on the **cobas c** 503 analytical unit.

Tina-quant Hemoglobin A1cDx Gen.3 offers two sample type specific applications, Hemolysate Application and Whole Blood Application, one for manually hemolyzed samples and one for whole blood samples respectively.

The Tina-quant Hemoglobin A1cDx Gen.3 assay first measures total hemoglobin (Hb) and glycated hemoglobin (HbA1c) in terms of either g/dL or mmol/L. Then the analyzer calculates the HbA1c/Hb ratio according to either IFCC or DCCT/NGSP. IFCC protocol reports the ratio in terms of mmol/mol HbA1c while the DCCT/NGSP protocol reports the ratio in terms of % HbA1c. Performance characteristics that support the measuring ranges claimed for Hb and HbA1c included limit of detection and linearity. These results were reported in terms of Hb and HbA1c individually. Patient sample values are reported in terms of the ratio of glycated to total hemoglobin. Method comparison, control recovery and precision were evaluated in terms of the ratio.

The following performance data were provided in support of the substantial equivalence determination:

4.1. Precision

4.1.1. Repeatability and Intermediate Precision

Precision measurements were conducted to evaluate repeatability (within-run precision) and intermediate precision (within-laboratory precision) according the CLSI guideline EP5-A3.

Two aliquots per sample were measured once each, in two runs per day, for 21 days, on 3 **cobas c** 503 analytical units and using 3 reagent lots per system. Ten total samples were evaluated in each run: two controls, PreciControl HbA1c norm and PreciControl HbA1c path, and eight human samples with approximate Hb1Ac concentrations of 4.9%, 6.6%, 7.3%, 8.2%, 12.5%, 14.6%, 12.3% and 13.1% for Whole Blood and 5.0%, 6.6%, 7.3%, 8.3%, 12.5%, 14.7%, 12.1% and 12.9% for Hemolysate.

The samples were randomized within each run. For each sample, the following was calculated: mean, repeatability and intermediate precision as CV and SD values and the upper 95% confidence interval for SD and CV values.

Table 2: Precision Results – Hemolysate Application, Analytical Unit 1

Mean % HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-lot		Intermediate Precision (total)	
	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)
Hem 1 4.96	0.025	0.5	0.006	0.1	0.071	1.4	0.016	0.3	0.077	1.6
Hem 2 6.62	0.027	0.4	0.013	0.2	0.053	0.8	0.059	0.9	0.085	1.3
Hem 3 7.32	0.035	0.5	0.000	0.0	0.053	0.7	0.067	0.9	0.092	1.3
Hem 4 8.32	0.039	0.5	0.009	0.1	0.056	0.7	0.083	1.0	0.108	1.3
Hem 5 12.54	0.057	0.5	0.011	0.1	0.100	0.8	0.203	1.6	0.234	1.9
HE_006 14.77	0.077	0.5	0.013	0.1	0.148	1.0	0.268	1.8	0.316	2.1
HE_007 12.14	0.055	0.5	0.023	0.2	0.100	0.8	0.181	1.5	0.215	1.8
HE_008 12.94	0.072	0.6	0.000	0.0	0.111	0.9	0.188	1.5	0.230	1.8
PCA1N 5.53	0.024	0.4	0.009	0.2	0.059	1.1	0.028	0.5	0.071	1.3
PCA1P 10.89	0.055	0.5	0.026	0.2	0.085	0.8	0.146	1.3	0.179	1.6

Table 3: Precision Results – Hemolysate Application, Analytical Unit 2

Mean % HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-lot		Intermediate Precision (total)	
	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)
Hem 1 4.96	0.027	0.5	0.005	0.1	0.034	0.7	0.015	0.3	0.046	0.9
Hem 2 6.59	0.035	0.5	0.000	0.0	0.038	0.6	0.057	0.9	0.077	1.2
Hem 3 7.29	0.041	0.6	0.000	0.0	0.043	0.6	0.068	0.9	0.090	1.2
Hem 4 8.28	0.039	0.5	0.015	0.2	0.046	0.6	0.093	1.1	0.112	1.4
Hem 5 12.43	0.069	0.6	0.027	0.2	0.038	0.3	0.175	1.4	0.193	1.6
HE_006 14.68	0.085	0.6	0.011	0.1	0.060	0.4	0.220	1.5	0.243	1.7
HE_007 12.05	0.063	0.5	0.018	0.1	0.036	0.3	0.163	1.4	0.179	1.5
HE_008 12.85	0.071	0.6	0.034	0.3	0.053	0.4	0.177	1.4	0.201	1.6
PCA1N 5.52	0.030	0.5	0.008	0.1	0.029	0.5	0.024	0.4	0.049	0.9
PCA1P 10.81	0.074	0.7	0.000	0.0	0.041	0.4	0.134	1.2	0.159	1.5

Table 4: Precision Results – Hemolysate Application, Analytical Unit 3

Mean % HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-lot		Intermediate Precision (total)	
	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)
Hem 1 4.94	0.027	0.5	0.010	0.2	0.031	0.6	0.023	0.5	0.048	1.0
Hem 2 6.57	0.030	0.5	0.004	0.1	0.035	0.5	0.049	0.7	0.067	1.0

Hem 3 7.28	0.032	0.4	0.000	0.0	0.038	0.5	0.054	0.7	0.073	1.0
Hem 4 8.28	0.040	0.5	0.000	0.0	0.041	0.5	0.070	0.8	0.091	1.1
Hem 5 12.43	0.063	0.5	0.024	0.2	0.045	0.4	0.162	1.3	0.181	1.5
HE_006 14.68	0.075	0.5	0.000	0.0	0.050	0.3	0.240	1.6	0.256	1.7
HE_007 12.07	0.064	0.5	0.000	0.0	0.039	0.3	0.146	1.2	0.164	1.4
HE_008 12.84	0.072	0.6	0.000	0.0	0.052	0.4	0.150	1.2	0.174	1.4
PCA1N 5.49	0.026	0.5	0.008	0.2	0.035	0.6	0.024	0.4	0.050	0.9
PCA1P 10.78	0.071	0.7	0.000	0.0	0.048	0.4	0.116	1.1	0.144	1.3

Table 5: Precision Results – Hemolysate Application, 3 Lots and 3 Analytical Units

Mean, %HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-Lot		Between-Device		Reproducibility (total)	
	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)
Hem 1, 4.96	0.026	0.5	0.007	0.1	0.049	1.0	0.019	0.4	0.009	0.2	0.059	1.2
Hem 2, 6.59	0.031	0.5	0.006	0.1	0.042	0.6	0.055	0.8	0.023	0.3	0.080	1.2
Hem 3, 7.30	0.036	0.5	0.000	0.0	0.045	0.6	0.063	0.9	0.019	0.3	0.088	1.2
Hem 4, 8.29	0.039	0.5	0.005	0.1	0.049	0.6	0.082	1.0	0.019	0.2	0.105	1.3
Hem 5, 12.47	0.063	0.5	0.022	0.2	0.070	0.6	0.179	1.4	0.061	0.5	0.212	1.7
HE_006, 14.71	0.079	0.5	0.010	0.1	0.098	0.7	0.242	1.6	0.053	0.4	0.278	1.9
HE_007, 12.08	0.061	0.5	0.016	0.1	0.067	0.6	0.163	1.3	0.048	0.4	0.193	1.6
HE_008, 12.88	0.072	0.6	0.017	0.1	0.078	0.6	0.172	1.3	0.055	0.4	0.210	1.6
PCA1N, 5.51	0.027	0.5	0.008	0.2	0.043	0.8	0.027	0.5	0.022	0.4	0.062	1.1
PCA1P, 10.83	0.067	0.6	0.000	0.0	0.062	0.6	0.132	1.2	0.054	0.5	0.169	1.6

Table 6: Precision Results – Whole Blood Application, Analytical Unit 1

Mean %HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-Lot		Intermediate Precision (total)	
	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)
WB 1, 4.87	0.038	0.8	0.000	0.0	0.043	0.9	0.045	0.9	0.073	1.5
WB 2, 6.60	0.026	0.4	0.015	0.2	0.030	0.4	0.071	1.1	0.083	1.3
WB 3, 7.37	0.032	0.4	0.018	0.2	0.032	0.4	0.084	1.1	0.097	1.3
WB 4, 8.24	0.039	0.5	0.008	0.1	0.048	0.6	0.089	1.1	0.108	1.3
WB 5, 12.59	0.056	0.4	0.025	0.2	0.061	0.5	0.154	1.2	0.177	1.4
WB_006, 14.69	0.079	0.5	0.043	0.3	0.067	0.5	0.168	1.1	0.202	1.4
WB_007, 12.34	0.062	0.5	0.032	0.3	0.057	0.5	0.152	1.2	0.176	1.4
WB_008, 13.14	0.061	0.5	0.015	0.1	0.055	0.4	0.165	1.3	0.185	1.4
PCA1N, 5.51	0.029	0.5	0.002	0.0	0.039	0.7	0.044	0.8	0.066	1.2
PCA1P, 11.20	0.055	0.5	0.011	0.1	0.057	0.5	0.129	1.2	0.152	1.4

Table 7: Precision Results – Whole Blood Application, Analytical Unit 2

Mean %HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-Lot		Intermediate Precision (total)	
	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)
WB 1, 4.88	0.032	0.7	0.013	0.3	0.026	0.5	0.031	0.6	0.053	1.1
WB 2, 6.58	0.031	0.5	0.010	0.2	0.025	0.4	0.077	1.2	0.087	1.3
WB 3, 7.35	0.040	0.5	0.000	0.0	0.029	0.4	0.091	1.2	0.103	1.4
WB 4, 8.21	0.042	0.5	0.011	0.1	0.035	0.4	0.106	1.3	0.120	1.5
WB 5, 12.53	0.070	0.6	0.029	0.2	0.047	0.4	0.164	1.3	0.186	1.5
WB_006, 14.62	0.095	0.6	0.035	0.2	0.091	0.6	0.198	1.4	0.240	1.6
WB_007, 12.25	0.078	0.6	0.000	0.0	0.035	0.3	0.176	1.4	0.195	1.6
WB_008, 13.06	0.073	0.6	0.009	0.1	0.052	0.4	0.163	1.2	0.186	1.4
PCA1N, 5.51	0.036	0.6	0.000	0.0	0.028	0.5	0.044	0.8	0.063	1.1
PCA1P, 11.13	0.062	0.6	0.033	0.3	0.043	0.4	0.162	1.5	0.182	1.6

Table 8: Precision Results – Whole Blood Application, Analytical Unit 3

Mean %HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-Lot		Intermediate Precision (total)	
	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)
WB 1, 4.86	0.029	0.6	0	0.0	0.035	0.7	0.044	0.9	0.064	1.3
WB 2, 6.55	0.031	0.5	0.009	0.1	0.025	0.4	0.080	1.2	0.090	1.4
WB 3, 7.31	0.039	0.5	0.006	0.1	0.026	0.4	0.090	1.2	0.101	1.4
WB 4, 8.17	0.043	0.5	0.012	0.2	0.036	0.4	0.096	1.2	0.111	1.4
WB 5, 12.51	0.079	0.6	0.000	0.0	0.064	0.5	0.154	1.2	0.184	1.5
WB_006, 14.62	0.082	0.6	0.000	0.0	0.107	0.7	0.191	1.3	0.234	1.6
WB_007, 12.23	0.068	0.6	0.028	0.2	0.056	0.5	0.157	1.3	0.182	1.5
WB_008, 13.05	0.083	0.6	0.000	0.0	0.064	0.5	0.162	1.2	0.193	1.5
PCA1N, 5.50	0.030	0.6	0.006	0.1	0.028	0.5	0.051	0.9	0.066	1.2
PCA1P, 11.10	0.071	0.6	0.000	0.0	0.043	0.4	0.137	1.2	0.160	1.4

Table 9: Precision Results – Whole Blood Application, 3 Lots and 3 Analytical Units

Mean %HbA1c	Repeatability (error)		Between-Run		Between-Day		Between-Lot		Between-Device		Reproducibility (total)	
	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)	SD	CV(%)
WB 1, 4.87	0.034	0.7	0.004	0.1	0.036	0.7	0.040	0.8	0.007	0.1	0.064	1.3
WB 2, 6.57	0.029	0.4	0.012	0.2	0.028	0.4	0.075	1.1	0.024	0.4	0.090	1.4
WB 3, 7.34	0.037	0.5	0.009	0.1	0.032	0.4	0.087	1.2	0.027	0.4	0.104	1.4
WB 4, 8.20	0.041	0.5	0.011	0.1	0.042	0.5	0.096	1.2	0.031	0.4	0.117	1.4
WB 5, 12.54	0.069	0.5	0.020	0.2	0.062	0.5	0.155	1.2	0.043	0.3	0.187	1.5
WB_006, 14.64	0.086	0.6	0.028	0.2	0.096	0.7	0.181	1.2	0.043	0.3	0.228	1.6
WB_007, 12.27	0.070	0.6	0.023	0.2	0.055	0.5	0.159	1.3	0.055	0.4	0.192	1.6
WB_008, 13.08	0.073	0.6	0.000	0.0	0.061	0.5	0.161	1.2	0.049	0.4	0.194	1.5
PCA1N, 5.51	0.032	0.6	0.002	0.0	0.032	0.6	0.047	0.8	0.007	0.1	0.065	1.2
PCA1P, 11.14	0.063	0.6	0.019	0.2	0.054	0.5	0.141	1.3	0.047	0.4	0.171	1.5

4.2. Analytical Sensitivity

4.2.1. Limit of Blank (LoB)

For determination of LoB, one analyte free sample was measured with three lots of Tina-quant Hemoglobin A1cDx Gen.3, in 10-fold determinations. Six runs were distributed over ≥ three days and measured using one **cobas c** 503 analytical unit. In total, 60 measurements were obtained per lot. Data analysis was based on determination of the 95th percentile of the 60 measured values. In our design (n=60) the 95th percentile is the average of the 57th and 58th value.

LoB was determined according to CLSI guideline EP17-A2.

Table 10: LoB Results

Hb LoB		**HbA1c LoB**	
mmol/L	**g/dL**	**mmol/L**	**g/dL**
0.0530 mmol/L	0.085 g/dL	0.0220 mmol/L	0.035 g/dL

This corresponds to a Limit of Blank of 15 mmol/mol (IFCC) and 3.5 % HbA1c (DCCT/NGSP) at a typical hemoglobin concentration of 13.2 g/dL (8.2 mmol/L).

4.2.2. Limit of Detection (LoD)

For determination of LoD, five unique human samples with low-analyte concentrations were measured with three lots of Tina-quant Hemoglobin A1cDx Gen.3 in two-fold determinations. The measurements were performed in six runs, over ≥ three days, on one **cobas c** 503 analytical unit. In total, 60 measurements were obtained per lot.

LoD is defined as the concentration, at which there is a 95% probability that a sample contains analyte.

LoD was determined according to CLSI guideline EP17-A2.

Table 11: LoD Results

Hb LoD		HbA1c LoD	
mmol/L	**g/dL**	**mmol/L**	**g/dL**
0.119 mmol/L	0.192 g/dL	0.0437 mmol/L	0.07 g/dL

This corresponds to a Limit of Detection of 22 mmol/mol (IFCC) and 4.2 % HbA1c (DCCT/NGSP) at a typical hemoglobin concentration of 13.2 g/dL (8.2 mmol/L).

4.3. Linearity/Assay Reportable Range

Separate dilution series, consisting of at least eleven levels, were prepared for each glycated hemoglobin (HbA1c) and total hemoglobin (Hb) using human hemolysate sample pools. The sample pools include HbA1c and Hb concentrations above the upper end of the corresponding measuring range. Hemolyzing reagent was used for the diluent. Samples were measured in triplicate and data analysis was performed separately for each sample.

The study was performed according to CLSI guideline EP6-A.

Table 12: Linearity Results according to CLSI EP6-A

Application	Analyte	Low End of Linear Range		High End of Linear Range	
		g/dL	**mmol/L**	**g/dL**	**mmol/L**
Hemolysate	Hb	3.04	1.89	40.4	25.1
	HbA1c	0.293	0.182	2.87	1.78

Table 13: Empirical First Order Regression Results

Application	Analyte	Slope	Intercept	Pearson's r
Hemolysate	Hb	1.019	-0.1552	0.9999
	HbA1c	0.991	-0.0026	0.9990

Linearity was determined throughout the claimed measuring range of:

Hb: 4 – 40 g/dL (2.48 – 24.8 mmol/L)

HbA1c: 0.3 – 2.6 g/dL (0.186 – 1.61 mmol/L)

This corresponds to a measuring range of 23-196 mmol/mol HbA1c (IFCC) and 4.2-20.1 % HbA1c (DCCT/NGSP) at a typical hemoglobin concentration of 13.2 g/dL (8.2 mmol/L).

4.4. Endogenous Interferences

A study evaluated several endogenous substances for potential interference with measurement of % HbA1c. The following nine endogenous substances were evaluated.

- Bilirubin
- Ditaurobilirubin
- Lipemia
- Rheumatoid Factors
- Total Protein
- Albumin
- Immunoglubulin (IgG)
- Glucose
- Triglycerides

Pooled whole blood samples, with two hemoglobin A1c levels, one near the medical decision level and one above it, were spiked with the maximum level of the above nine interferents, in separate preparations, resulting in eighteen spiked samples. These samples were then hemolyzed

with Tina-quant HbA1c Hemolyzing Reagent. Another pool, without interferent, was equally hemolyzed. A ≥ ten-level dilution series was created for each of the eighteen spiked samples, using the interferent-free pools as the diluent.

The eighteen dilution series were tested in ten-fold, using one reagent lot, one **cobas c** 503 analytical unit, in a single run and within one calibration cycle. Additionally, PreciControl HbA1c norm and PreciControl HbA1c path were used as the controls. The mean of the ten replicates was compared to the result from the reference sample (aliquot with no interferent). The comparison was evaluated as a percent deviation. For purposes of this experiment, the data was collected using the Hemolysate Application and was representative of both Hemolysate and Whole Blood Applications.

Table 14: Endogenous Interference

Potential Interferent	Claimed Maximum Concentration without Interference
Bilirubin	60 mg/dL
Ditaurobilirubin	60 mg/dL
Lipemia	400 mg/dL
Rheumatoid Factors	750 IU/mL
Total Protein	21 g/dL
Albumin	60 g/L
Immunoglobulin (IgG)	60 g/L
Glucose	1000 mg/dL
Triglycerides	1584 mg/dL

4.5. Cross-Reactivity

This study was conducted to evaluate the Tina-quant Hemoglobin A1cDx Gen.3 assay on the **cobas c** 503 analytical unit for potential cross-reactivity with the following hemoglobin fractions and glycated albumin.

- HbA0
- HbA1(a+b)
- Labile HbA1c
- Carbamylated Hb
- Acetylated Hb
- Glycated Albumin

A series of experiments were performed using one reagent lot, on one **cobas c** 503 analytical unit, in a single run, within one calibration cycle. PreciControl HbA1c norm and PreciControl HbA1c path were used as the controls. Ten replicates of each sample were analyzed for each dilution level. The median % HbA1c of each dilution level was compared to the median % HbA1c from dilution level zero (without cross-reactant). For purposes of this experiment, the data was collected using the Hemolysate Application and was representative of both Hemolysate and Whole Blood Applications.

Table 15: Cross-Reactivity

Cross-Reactant		Max Whole Blood Cross-Reactant Concentration	Max Whole Blood Cross-Reactant Concentration with no Interference
HbA0	HbA1c Level 1	120 g/dL	120 g/dL
	HbA1c Level 2	120 g/dL	120 g/dL
HbA1(a+b)	HbA1c Level 1	1.6 g/dL	0.96 g/dL
	HbA1c Level 2	1.6 g/dL	1.6 g/dL
Carbamylated Hb	HbA1c Level 1	2.0 g/dL	2.0 g/dL
	HbA1c Level 2	2.0 g/dL	2.0 g/dL
Acetylated Hb	HbA1c Level 1	2.0 g/dL	2.0 g/dL
	HbA1c Level 2	2.0 g/dL	2.0 g/dL
Glycated Albumin	HbA1c Level 1	10 g/dL	10 g/dL
	HbA1c Level 2	10 g/dL	10 g/dL
Labile HbA1c	HbA1c Level 1	1000 mg/dL	1000 mg/dL
	HbA1c Level 2	1000 mg/dL	1000 mg/dL

4.6. Hemoglobin Variants

Hemoglobin variant testing was conducted to determine if significant interference with any of the major hemoglobin variants occurred when using the Tina-quant Hemoglobin A1cDx Gen.3 assay on **cobas c** 503 analytical unit. Hemoglobin variants are structurally altered hemoglobin molecules with at least one amino acid exchange, compared to the normal beta chain of hemoglobin. These changes are caused by mutations in the coding region of the globin genes which encode the protein part of hemoglobin. The most common hemoglobin variants are HbS, HbC, HbD and HbE. Additionally, in some conditions fetal hemoglobin, HbF, is elevated. Also, the erythrocytes of some patients (e.g. beta thalassemia minor) contain elevated levels of HbA2.

Therefore, it is crucial to ensure accurate HbA1c results from patients who are carriers of these variants.

Table 16: Hemoglobin Variant Samples

Variant Type	Number of Samples	% Variant	HbA1c %
HbS	30	35-41% S	4.35 - 12.7
HbC	30	28-37% C	4.90 - 14.1
HbE	30	24-27% E	5.17 - 10.0
HbD	29	36-42% D	5.17 - 9.70
HbA2	15	4.3-6.5% A2	5.10 - 9.80
Elevated HbF	19	3.2-39% F	6.10 -9.30

Each sample was tested once, in at least one run, on one **cobas c** 503 analytical unit. Results obtained with the Tina-quant Hemoglobin A1cDx Gen.3 assay on the **cobas c** 503 analytical unit were compared to those obtained with the corresponding reference method. For purposes of this experiment, the data was collected using the Hemolysate Application and was representative of both Hemolysate and Whole Blood Applications.

Table 17: Hemoglobin Variant Testing

Percent Relative Bias from Reference Method at Low and High Concentrations of HbA1c Samples				
HbVariant	Around 6.5% HbA1c		Around 9% HbA1c	
	Relative % Difference	Range	Relative % Difference	Range
HbS	-2.5	-7.2 – 3.2	-4.0	-9.3 – (-2.0)
HbC	-3.9	-7.7 – 2.8	-6.0	-4.6 – (-3.6)
HbE	-0.1	-5.5 – 5.7	-1.2	-5.2 – 0.6
HbD	-1.8	-4.5 – 3.0	-2.6	-3.3 – 0.2
HbA2	-1.0	-4.1 – 2.7	0.4	-2.2 – 1.1
HbF	Specimens containing high amounts of HbF (>7%) may yield lower than expected HbA1c values.			

4.7. Exogenous Interferences – Drugs

The purpose of this study was to evaluate drugs for potential interference with the Tina-quant Hemoglobin A1cDx Gen.3 assay measured on the **cobas c** 503 analytical unit.

The eighteen commonly used drugs listed below were added to samples and examined for potential effect on % HbA1c determination. Drug interference testing was performed with

hemolysate samples at two different HbA1c levels, approximately 6% and 9% HbA1c. Each drug was added in two defined concentrations with concentration one being several times (typically five times) the maximum daily dosage and concentration 2 being the maximum daily dosage level. Concentration one was performed for screening purposes only and concentration two was the relevant drug concentration for determining interferences with the assay. Samples were measured in ten-fold using the **cobas c** 503 analytical unit. The median value was compared to the reference value (HbA1c sample with no drug added) and the deviation from the reference was calculated. Drug interference studies were conducted using the Hemolysate Application and were representative of both Hemolysate and Whole Blood Applications.

- N-Acetylcysteine
- Ampicillin-Na
- Ascorbic acid
- Cefoxitin
- Heparin
- Levodopa
- Methyldopa + 1.5
- Metronidazole
- Doxycyclin
- Acetylsalicylic acid
- Rifampicin
- Gammagard
- Cyclosporine
- Phenylbutazone
- Acetaminophen
- Ibuprofen
- Theophylline
- Tolbutamide

4.8. Sample Matrix Comparison

The purpose of this study was to evaluate Hemoglobin A1c determination, with the Tina-quant Hemoglobin A1cDx Gen.3 assay, in the presence of anticoagulants.

At least 40 samples of each anticoagulant and at least 40 half-filled tubes of each anticoagulant were evaluated. The filled and corresponding half-filled (double concentrated) sample tubes were from one donor. Matrix comparison studies were conducted using the Hemolysate Application and was representative of both Hemolysate and Whole Blood Applications.

Table 18: Matrix Comparison Results

Sample Type	Anticoagulant	Tube Fill	Mean Difference	Upper 95%	Lower 95%
Hemolysate	K_2-EDTA	½ Full	0.004	0.124	-0.116
	K_3-EDTA	Full	-0.005	0.104	-0.114
		½ Full	0.004	0.140	-0.131
	Na Heparin	Full	0.000	0.120	-0.121
		½ Full	-0.026	0.129	-0.181
	Li Heparin	Full	-0.003	0.081	-0.088
		½ Full	-0.015	0.138	-0.167
	NaF/Potassium oxalate	Full	0.009	0.123	-0.105
		½ Full	0.019	0.135	-0.097
	EDTA/Fluoride	Full	0.003	0.100	-0.094
		½ Full	0.017	0.172	-0.139

4.9. Method Comparison

A method comparison study was performed to compare the sample results from the candidate method, Hemoglobin A1cDx Gen.3 assay on the **cobas c** 503 analytical unit, to results from Tosoh Automated Glycohemoglobin Analyzer HLC-723G8 (Tosoh), the NGSP method. This study was conducted with both the Tina-quant Hemoglobin A1cDx Gen.3 Whole Blood Application and Hemolysate Application.

One hundred and seventy-one whole blood samples and one hundred seventy-three hemolysate samples from the secondary NGSP reference laboratory were used in the evaluation. These samples were measured by the secondary NGSP reference laboratory using the Tosoh HPLC system (X-axis) and the Roche Tina-quant Hemoglobin A1cDx Gen.3 test system (Y-axis). The samples were tested over a 3-day period, with one lot of reagent, on one **cobas c** 503 analytical unit.

Table 19: Method Comparison Sample Distribution

% HbA1c	# Samples Tested		% Samples Tested	
	Whole Blood	Hemolysate	Whole Blood	Hemolysate
≤ 5%	6	6	3.5%	3.5%
5-6 %	23	24	13.5%	13.9%

>6 – 6.5%	31	32	18.1%	18.5%
>6.5 – 7%	37	37	21.6%	21.4%
>7 – 8%	25	25	14.6%	14.5%
>8 – 9%	13	13	7.6%	7.5%
>9%	36	36	21.1%	20.8%
Total	171	173	100%	100%

The method comparison demonstrated good agreement between Roche Tina-quant Hemoglobin A1cDx Gen.3 assay and the NGSP Tosoh reference method. The tables below summarize the bias between the Tina-quant Hemoglobin A1cDx Gen.3 assay and the NGSP Tosoh reference method.

Table 20: Difference Plot Analysis Data Summary

	Whole Blood Application	**Hemolysate Application**
Mean bias vs. NGSP TOSOH	-0.046%	0.046%
Mean bias at lower 95% CI	-0.410%	-0.338%
Mean bias at upper 95% CI	0.318%	0.431%

Table 21: Bias at Concentration Data Summary

% HbA1c	**% Relative Bias Whole Blood Application**	**% Relative Bias Hemolysate Application**
5%	-2.4%	0.6%
6.5%	-1.2%	0.8%
8%	-0.4%	1.0%
12%	0.7%	1.2%

4.10. Total Error

Using the results of bias estimation (%Bias) generated during the method comparison study and precision estimates (%CV) from the precision study, the Total Error (TE) was calculated using the following equation:

%TE = |%Bias| + 1.96 * %CV * (1 + %Bias/100).

Table 22: Total Error – Hemolysate Application

%HbA1c	% BIAS	Precision (%CV)	Total Error (%)
4.96	0.63	1.2	3.0
6.59	0.87	1.2	3.3
8.29	1.02	1.3	3.5
12.1	1.20	1.6	4.4

Table 23: Total Error – Whole Blood Application

%HbA1c	% BIAS	Precision (%CV)	Total Error (%)
4.87	-2.56	1.3	5.2
6.57	-1.12	1.4	3.8
8.20	-0.30	1.4	3.1
12.3	0.79	1.6	3.9

5. ADDITIONAL INFORMATION

5.1. Other Devices Marketed with This Assay

The following devices are required, but not provided:

- C.f.a.s. (Calibrator for automated systems) HbA1c, k052101
- PreciControl HbA1c norm, k103099
- PreciControl HbA1c path, k103099

6. CONCLUSIONS

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