

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

A. 510(k) Number:

k041764

B. Purpose For Submission:

Premarket Notification for Ortho-Clinical Diagnostics, Inc., to manufacture and market the VITROS® Chemistry Products %A1c Reagent Kit, VITROS Chemistry Products Calibrator Kit 18, VITROS Chemistry Products FS Calibrator 1, and the VITROS Chemistry Products %A1c Performance Verifiers I and II.

C. Analyte:

Glycosylated hemoglobin assay and controls

D. Type of Test:

Quantitative turbidimetric, inhibition immunoassay

E. Applicant:

Ortho Clinical Diagnostics

F. Proprietary and Established Names:

VITROS Chemistry Products %A1c Reagent Kit
VITROS Chemistry Products Hemolyzing Reagent
VITROS Chemistry Products Calibrator Kit 18
VITROS Chemistry Products FS Calibrator 1
VITROS Chemistry Products %A1c Performance Verifiers I and II

G. Regulatory Information:

1. Regulation section:

21 CFR §864.7470, Glycosylated hemoglobin assay

21 CFR §862.1660, Single (specified) analyte controls (assayed and unassayed)

21 CFR §862.1150, Calibrator, Secondary

2. Classification:

Class II (assay)

Class I (controls)

Class II (calibrator)

3. Product Code:

LCP (assay)

JJX (controls)

JIT (calibrator)

4. Panel:
81 (Hematology)
75 (Chemistry)
75 (Chemistry)

H. Intended use(s):

1. Intended use(s)

VITROS Chemistry Products %A1c Reagent Kit: For in vitro diagnostic use only. VITROS Chemistry Products %A1c is used to calculate percent glycated hemoglobin (%A1c) in pre-treated human whole blood by quantitative measurement of hemoglobin (Hb) and hemoglobin A1c (HbA1c). Measurements of percentage A1c are effective in monitoring long-term glucose control in individuals with diabetes mellitus.

VITROS Chemistry Products Calibrator Kit 18: For in vitro diagnostic use only. VITROS Chemistry Products Calibrator Kit 18 is used in conjunction with VITROS Chemistry Products FS Calibrator 1 to calibrate VITROS 5, 1 FS Chemistry Systems for the calculation of percent glycated hemoglobin (%A1c).

VITROS Chemistry Products %A1c Performance Verifier I and II: For in vitro diagnostic use only. VITROS Chemistry Products %A1c Performance Verifiers are assayed controls used to monitor performance of %A1c Reagent Kit on VITROS 5, 1 FS Chemistry Systems.

2. Indication(s) for use:

VITROS Chemistry Products %A1c Reagent Kit: For in vitro diagnostic use only. VITROS Chemistry Products %A1c is used to calculate percent glycated hemoglobin (%A1c) in pre-treated human whole blood by quantitative measurement of hemoglobin (Hb) and hemoglobin A1c (HbA1c). Measurements of percentage A1c are effective in monitoring long-term glucose control in individuals with diabetes mellitus.

VITROS Chemistry Products Calibrator Kit 18: For in vitro diagnostic use only. VITROS Chemistry Products Calibrator Kit 18 is used in conjunction with VITROS Chemistry Products FS Calibrator 1 to calibrate VITROS 5, 1 FS Chemistry Systems for the calculation of percent glycated hemoglobin (%A1c).

VITROS Chemistry Products %A1c Performance Verifier I and II: For in vitro diagnostic use only. VITROS Chemistry Products %A1c Performance Verifiers are assayed controls used to monitor performance of %A1c Reagent Kit on VITROS 5, 1 FS Chemistry Systems.

3. Special condition for use statement(s):
For Prescription Use.
4. Special instrument Requirements:
VITROS 5, 1 FS Chemistry Systems

I. Device Description:

The VITROS 5, 1 FS Chemistry System is a fully automated clinical chemistry analyzer intended for use in the in vitro determination of various analytes in human specimens (serum, plasma, urine, and cerebrospinal fluid). The VITROS 5, 1 FS Chemistry System is designed for use with VITROS Chemistry Products MicroTip and Thin Film assays.

The system is comprised of four main elements:

1. The VITROS 5, 1 FS Chemistry System-instrumentation, which provides automated use of chemistry reagents. The VITROS 5, 1 FS Chemistry System was cleared for market by a separate 510(k) premarket notification (k031924).
2. The VITROS Chemistry Products range of MicroTip assays, in this case the VITROS Chemistry Products %A1c Reagent Kit, VITROS Chemistry Products Hemolyzing Reagent, VITROS Chemistry Products Calibrator Kit 18, VITROS Chemistry Products FS Calibrator1, and the VITROS Chemistry Products %A1c Performance Verifiers, which are combined by the VITROS 5, 1 FS Chemistry System to perform the VITROS %A1c assay.
3. The VITROS Chemistry Products Thin Film range of products, which are dry, multilayered, analytical elements, coated on polyester supports. The thin film products each have their own 510(k) clearance numbers and were cleared for market for use on the VITROS 5, 1 FS Chemistry System through submission of information required by the ODE Guidance Document: "Data For Commercialization Of Original Equipment Manufacturer, Secondary and Generic Reagents for Automated Analyzers". The required information was provided in the VITROS 5, 1 FS Chemistry System premarket notification (k031924)
4. Common reagents used by multiple assays on the VITROS System (in this case, VITROS Chemistry Products FS Diluent Pack 2 and VITROS Chemistry Products FS Reconstitution Diluent).

J. Substantial Equivalence Information:

1. Predicate device name(s):
Tosoh Medics, Inc. A1c 2.2 Plus Glycohemoglobin Assay
2. Predicate k number(s):
k972265
3. Comparison with Predicate:

The VITROS Chemistry Products %A1c Reagent Kit, VITROS Chemistry Products Calibrator Kit 18 and VITROS Chemistry Products FS Calibrator 1 are substantially equivalent to the A1c 2.2 Plus Glycohemoglobin Assay (Tosoh Medics, Inc.), which was cleared by the FDA (k972265) for IVD use.

The relationship between the VITROS %A1c assay and the predicate device in National Glycohemoglobin Standardization Program (NGSP) units, determined by least squares linear regression, is:

$$\text{VITROS \%A1c assay} = 1.0374X - 0.3426 \%A1c$$

With a correlation coefficient of 0.9872,

where X is the Tosoh A1c 2.2 Plus Glycohemoglobin Assay on the TOSOH Automated Glycohemoglobin Analyzer.

In addition to the above mentioned correlation study, studies were performed to determine the precision, expected values, linearity and specificity of the VITROS %A1c assay. The table below lists the characteristics of the VITROS %A1c Assay and the A1c 2.2 Plus Glycohemoglobin Assay (Tosoh Medics, Inc.).

Comparison to predicate device: Reagent Kit

Device Characteristics	VITROS %A1c Assay (New Device)	A1c 2.2 Plus Glycohemoglobin Assay (Predicate device)
Intended Use	To determine %A1c in whole blood	To determine %A1c in whole blood
Standardization	Traceable to both the Diabetes Control and Complications Trial (DCCT) and IFCC reference methods. Certified by the National Glycohemoglobin Standardization Program (NGSP)	Traceable to both the Diabetes Control and Complications Trial (DCCT) and IFCC reference methods. Certified by the National Glycohemoglobin Standardization Program (NGSP)
Basic Principle	Turbidimetric inhibition immunoassay	High performance liquid chromatography (HPLC)
Reportable Range	4-14% A1c (NGSP)	4-19% A1c (NGSP)
Reagents	Liquid ready to use	Non-porous cation exchange column
Instrumentation	VITROS 5, 1 FS Chemistry System	A1c 2.2 Plus Automated Glycohemoglobin Analyzer (Tosoh Medics, Inc.)
Sample Type	Whole blood (EDTA, heparin, sodium fluoride potassium oxalate)	Whole blood (undiluted or diluted)

The table below lists the characteristics of the VITROS Chemistry Products %A1c Performance Verifiers and the VITROS Chemistry Products Performance Verifiers.

Comparison to predicate device: Performance Verifiers

Device Characteristics	VITROS %A1c Performance Verifiers (New Device)	VITROS Performance Verifiers (Predicate device)
Intended Use	VITROS %A1c Performance Verifier is an assayed control used to monitor performance of %A1c Reagent Kit on VITROS 5, 1 FS Chemistry Systems.	VITROS Performance Verifier is an assayed control used to monitor performance on VITROS Chemistry Systems.
Matrix	A base matrix of freeze-dried hemolysate derived from human and ovine blood to which surfactants, stabilizer, and preservatives have been added.	A base matrix of freeze-dried human serum to which enzymes, electrolytes, stabilizers, preservatives and other organic analytes have been added.
Levels	Low and High	Low and High

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

Results are traceable to both the Diabetes Control and Complications Trial (DCCT) reference method and the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Reference Method for the measurement of HbA1c in human blood.

Certified by the National Glycohemoglobin Standardization Program (NGSP)

NCCLS Protocol EP5-A - Evaluation of Precision Performance of Clinical Chemistry Devices.

NCCLS EP6-A Evaluation of the Linearity of Quantitative Analytical Methods.

NCCLS EP7-A Interference Testing in Clinical Chemistry

EP9-A2 Method Comparison and Bias Estimation Using Patient Samples.

**L. Test Principle:
Hemoglobin A1c**

The determination of % glycated hemoglobin (%A1c) is performed using the VITROS Chemistry Products %A1c Reagent Kit in conjunction with the VITROS Chemistry

Products Calibrator Kit 18 and the VITROS Chemistry Products FS Calibrator 1 on the VITROS 5,1 FS Chemistry System. The VITROS Chemistry Products %A1c Reagents are two dual chambered packages containing anti-HbA1c antibody to form a soluble antigen-antibody complex. Unbound anti-HbA1c antibody reacts with polyhaptan (hexapeptide-glycan, Reagent 2) to form an insoluble antibody-polyhaptan immune complex, which is measured turbidimetrically at 340 nm. Once a calibration has been performed for each reagent lot, the hemoglobin A1c concentration in each unknown sample can be determined using the stored calibration curve and the measured absorbance obtained in the assay of the sample.

Hemoglobin

The hemoglobin concentration is determined by a separate assay protocol. Hemoglobin in the hemolyzed whole blood sample is converted in an alkaline solution to a hematin derivative that is measured bichromatically at 575 nm and 700 nm. Once a calibration has been performed for each reagent lot, the hemoglobin concentration in each unknown sample can be determined using the stored calibration curve and the measured absorbance obtained in the assay of the sample.

%A1c

The %A1c is calculated from the quantitative measurements of hemoglobin and hemoglobin A1c.

Reaction Sequence

HbA1c Step 1: Immune Complex HbA1c

	R1	
Hemolyzed whole blood	----->	HbA1c-anti-HbA1c immune complex
	Excess anti-HbA1c	and unbound anti-HbA1c (soluble)

HbA1c Step 2: Detect unbound (free) anti-HbA1c

	R2	
Unbound anti-HbA1c	----->	HbA1c polyhaptan/anti-HbA1c immune
	Excess HbA1c polyhaptan	complex (turbid/ insoluble)

Hemoglobin

	R1 (Hb)	
Hemolyzed whole blood	----->	hemoglobin Chromophore
	Reagent	

M. Performance Characteristics (if/when applicable):

1. Analytical performance:
 - a. *Precision/Reproducibility:*

Precision was evaluated with quality control materials on the VITROS 5, 1 FS Chemistry System following NCCLS Protocol EP5-A - Evaluation of Precision Performance of Clinical Chemistry Devices.

Two runs on two VITROS 5, 1 FS Systems with two reagent lots were performed on 20 different days. Runs within day were separated by at least two hours. Each run consisted of two control fluids and two patient sample pools assayed in duplicate to evaluate system precision. The calibrator samples were used to obtain a calibration for each week of the experiment. This process introduced the variability inherent in the act of calibration.

The results of the test samples were analyzed by ANOVA as detailed in NCCLS EP5-A. A nested ANOVA was performed to within-day, day-to-day, week-to-week (cal-to-cal) and total within-lab precision. The ANOVA results represent an approximation of the total, within-laboratory variability that would be observed using a single lot of reagents.

The within-day and within-laboratory precision is summarized in the table below.

Precision for %A1c: Whole Blood

System	NGSP				IFCC				N	Days
	Mean Value % A1c	With-in Day SD*	With-in Lab SD**	With-in Lab CV%	Mean Value %A1c	With-in Day SD*	With-in Lab SD**	With-in Lab CV%		
VITROS 5,1 FS	5.3	0.05	0.06	1.13	3.4	0.05	0.06	1.88	80	20
	6.3	0.05	0.08	1.33	4.6	0.05	0.09	2.01	80	20
	9.6	0.11	0.20	2.05	8.2	0.11	0.22	2.64	80	20
	11.3	0.12	0.19	1.66	10.0	0.13	0.20	2.05	80	20

* Within Day precision was determined using two runs/ day with two replications per run.

** Within Lab precision was determined using a single lot of reagents on a single analyzer, calibrating once a week.

b. Linearity/assay reportable range:

The evaluation was performed according to NCCLS EP6-A Evaluation of the Linearity of Quantitative Analytical Methods.

VITROS % A1c assay results are derived from the independently measured hemoglobin (Hb) and glycated hemoglobin (HbA1c) assay components. The linearity and reportable range of the %A1c assay is based on the linearity of the Hb and HbA1c assay components.

To estimate the linearity of the hemoglobin assay component, a high pool was created by reconstituting the highest calibrator to give a hemoglobin value >30

g/dL. The low pool, saline, had a concentration of 0 g/dL. The high pool was diluted with the low to create 9 additional pools of intermediate concentrations, for a total of 11 evaluation fluids.

The linearity of the HbA1c assay component and the %A1c assay were estimated in two steps: high-end and low-end linearity. To establish the high-end linearity, the lowest and the highest analyte containing calibrators were used as the low and high pools. The high pool was diluted with the low pool to create 9 additional pools of intermediate concentrations, for a total of 11 evaluation fluids. To estimate the low-end linearity, the lowest analyte containing calibrator was used as the high pool. A low pool was created by diluting the high pool 10-fold with FS Reconstitution Diluent (processed water). Admixtures of the low and high pools were made producing additional pools of intermediate concentrations, for a total of 11 evaluation fluids. Linearity of the %A1c assay was examined at normal (~ 14 g/dL) hemoglobin concentration.

Four replicates of each linearity fluid level and VITROS Chemistry Products % A1c Performance Verifiers I and II were tested on the VITROS 5,1 FS Chemistry System. This experiment was performed with two lots of calibrators and matched reagents. The VITROS 5, 1 FS Chemistry System was operated within normal conditions and was calibrated for the appropriate lot.

The data were screened for outliers according to the statistical outlier test in NCCLS EP5-A. Three replicate outliers were excluded in the analysis. For each reagent lot, the system responses were plotted against the percent high pool and against the calculated values to assess the degree to which the plotted curve conforms to a straight line. Acceptable linearity was defined as when the bias between predicated and calculated %A1c values was within predetermined acceptance limits.

The data supports a linear range of 4 to 14 %A1c (NGSP units) and 2 to 13% A1c (IFCC units) over the hemoglobin range of 6 to 22 g/dL and HbA1c range of 0.2 to 2.5 g/dL for the VITROS Chemistry Products %A1c assay when run on the VITROS 5,1 FS Chemistry System.

c. Traceability (controls, calibrators, or method):

Values assigned to the VITROS Chemistry Products Calibrator Kit 18 and VITROS Chemistry Products FS Calibrator Kit 1 for %A1c is traceable to the IFCC (international Federation of Clinical Chemistry and Laboratory Medicine) Reference Method.¹ The derived result (%A1c) is calculated from the individual quantitative results for hemoglobin (Hb) and glycated hemoglobin (HbA1c). NGSP results are derived from the International Federation of Clinical Chemistry (IFCC) %A1c units using the Master Equation¹⁴, $NGSP = (0.915 \times IFCC) + 2.15$.

¹ Jeppsson, Jan-Olof, et al, IFCC Scientific Working Group on HbA1c Standardization and Network of Reference Laboratories for HbA1c. Approved IFCC Reference Method for the Measurement of HbA1c Clin Chem Lab Med 2002; 40(1): 78-89.

¹⁴ Hoelzel W, Weykamp C, Jeppsson J, Miedema K, Barr JR, Goodall I, Hoshino T, John G, Kobold U, Little R, Mosca A, Mauri P, Paroni R, Susanto F, Takei I, Theinpont L, Umemoto M, Weidmeyer H. IFCC Working Group on HbA1c Standardization, IFCC Reference System for Measurement of Hemoglobin A1c in Human Blood and the National Standardization Schemes in the United States, Japan and Sweden: A Method Comparison Study, Clin Chem 2004 50 (1): 166-174.

d. Detection limit:

See linearity/assay reportable range in *b.* above.

e. Analytical specificity:

Cross-reactants:

Labile Glycated Hemoglobin: Test samples were prepared by spiking whole blood samples with glucose stock solution to a final glucose concentration of 1400 mg/dL in each sample. Control samples were prepared by spiking whole blood samples with purified water. The samples were incubated at 37°C for 4 hours and were tested for %A1c. Three determinations of each test substance were made using two reagent lots on one VITROS 5, 1 FS Chemistry System.

Performance Data

The mean %A1c value, SD and CV (%) were calculated for each control pool and test substance pool. The bias and the percent bias (also called percent interference) are calculated as shown below:

Bias = (Mean %A1c of test substance pool) – (Mean % A1c of control pool)

% Bias = $\frac{(\text{Mean of \%A1c of test substance pool}) - (\text{Mean \% A1c of control pool})}{\text{Mean \% A1c of control pool}} \times 100$

Mean % A1c of control pool

The % bias was compared to acceptance criteria.

Labile Glycated Hemoglobin Test Results:

Reagent Lot 91-5397									
Controls				Test			Specificity		
Sample ID	% A1c	STD DEV	%CV	%A1c	STD DEV	%CV	Bias	%Bias	(± 9.0%)
Sample 1	5.72	0.03	0.46	5.77	0.05	0.82	0.05	0.90	Pass
Sample 2	5.95	0.04	0.69	5.93	0.04	0.63	-0.02	-0.40	Pass
Sample 3	10.41	0.18	1.70	10.38	0.07	0.70	-0.04	-0.34	Pass
Sample 4	11.36	0.11	1.00	11.54	0.20	1.69	0.18	1.59	Pass
Reagent Lot 92-5581									
Controls				Test			Specificity		
Sample ID	% A1c	STD DEV	%CV	%A1c	STD DEV	%CV	Bias	%Bias	(± 9.0%)
Sample 1	5.85	0.05	0.89	5.89	0.05	0.93	0.01	0.18	Pass
Sample 2	6.10	0.06	1.01	6.10	0.06	0.95	0.00	-0.03	Pass
Sample 3	10.00	0.18	1.76	10.02	0.13	1.25	0.03	0.29	Pass
Sample 4	10.81	0.32	3.00	11.14	0.21	1.92	0.33	3.05	Pass

This method is unaffected by the presence of labile glycated hemoglobin.

Hemoglobin Variants:

Eighteen (18) whole blood samples containing hemoglobin variants (S, C, E and F) were acquired. The %A1c values for these samples were obtained from the Diabetes Diagnostics Laboratory using the Primus A1c HPLC method. Three determinations of each test substance were made using two reagent lots on one VITROS 5, 1 FS Chemistry System. The mean %A1c value, SD and CV% were calculated for each control pool and test substance pool. The bias was calculated (Bias = VITROS %A1c – Primus HPLC %A1c) and compared to acceptance criteria.

Samples Containing Hemoglobin S

Abnormal Hb Type	Primus A1c %	91-5397 %A1c	92-5581 %A1c	91-5397 %A1c Bias	92-5581 % Bias	Specificity Goal 91-5397	Specificity Goal 92-5581
HbS	5.4	5.33	5.39	-0.07	-0.01	Pass	Pass
HbS	5.5	5.51	5.53	0.01	0.03	Pass	Pass
HbS	5.8	5.73	5.68	-0.07	-0.12	Pass	Pass
HbS	6	5.67	5.75	-0.33	-0.25	Pass	Pass
HbS	6.3	5.99	6.00	-0.31	-0.30	Pass	Pass
HbS	6.4	6.23	6.36	-0.17	-0.04	Pass	Pass
HbS	6.6	6.41	6.43	-0.19	-0.17	Pass	Pass
HbS	6.7	6.18	6.21	-0.52	-0.49	Pass	Pass

HbS	7.5	7.01	7.06	-0.49	-0.44	Pass	Pass
HbS	10.1	9.22	9.28	-0.88	-0.82	Pass	Pass
HbS	10.7	10.96	11.58	0.26	0.88	Pass	Pass
HbS	13.4	12.66	12.57	-0.74	-0.83	Pass	Pass
Mean Bias				-0.29	-0.21		

Samples Containing Hemoglobin C

HbC	6	5.54	5.62	-0.46	-0.38	Pass	Pass
HbC	6.2	6.49	6.46	0.29	0.26	Pass	Pass
HbC	6.3	5.75	5.76	-0.55	-0.54	Pass	Pass
HbC	10.5	10.82	11.29	0.32	0.79	Pass	Pass
Mean Bias				-0.10	0.03		

Samples Containing Hemoglobin C and F

HbC, F	4.8	5.17	5.14	0.37	0.34	Pass	Pass
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Samples Containing Hemoglobin E

HbE	6.5	6.09	6.09	-0.41	-0.41	Pass	Pass
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The reagent supplier tested HbA0, HbA1a, HbA1b, HbA1a1, HbA1a2, acetylated hemoglobin, carbamylated hemoglobin, and glycated albumin with the VITROS Chemistry Products %A1c reagents.

Interferents:

Testing for ascorbic acid, bilirubin, and intralipid followed the protocol outlined in NCCLS EP7-A Interference Testing in Clinical Chemistry for paired-difference method. Two whole blood sample pools, one containing %A1c level in the normal range (4-6%) and another containing elevated % A1c (>9.0%) were used as base pools. Solutions of test substances were prepared by adding various levels of the substance to the base pools. Samples were prepared and tested on the same day. Three determinations of each test substance were made using two reagent lots on one VITROS 5, 1 FS Chemistry System.

The reagent supplier tested Acetaminophen, Acetylcysteine, Acetylsalicylic Acid, Ampicillin, Ascorbic Acid, Calcium-Dobesilate, Cefoxitin, Cyclosporin, Doxycycline, Ibuprofen, Intralipid, Levodopa, Methyldopa, Metronidazole, Phenylbutazone, Rifampicin, Rheumatoid factor, and Theophylline with the VITROS Chemistry Products %A1c reagents.

Data provided by the reagent supplier indicates that Rheumatoid factor (RF) up to 750 IU/mL does not interfere.

The substances listed in the table below were tested with the VITROS Chemistry Products % A1c reagents at % A1c values of approximately 5.0

% to 7.0% and found not to interfere, bias < 0.5%, at the concentration shown.

Substance*	Concentration	
Acetaminophen	20 mg/dL	1.32 mmol/L
Acetylcysteine	15 mg/dL	0.92 mmol/L
Acetylsalicylic acid	100 mg/dL	5.55 mmol/L
Ampicillin	100 mg/dL	2.86 mmol/L
Ascorbic acid	80 mg/dL	45.42 mmol/L
Bilirubin	50 mg/dL	0.86 mmol/L
Ca-dobesilate	20 mg/dL	0.46 mmol/L
Cefoxitin	250 mg/dL	5.56 mmol/L
Cyclosporin	0.5 mg/dL	4 µmol/L
Doxycycline	5 mg/dL	0.1 mmol/L
Ibuprophen	50 mg/dL	2.24 mmol/L
Intralipid	1000 mg/dL	Not Applicable
Levodopa	2 mg/dL	0.1 mmol/L
Methyldopa	2 mg/dL	90 µmol/L
Metronidazole	20 mg/dL	1.17 mmol/L
Phenylbutazone	40 mg/dL	1.3 mmol/L
Rifampicin	6 mg/dL	70 µmol/L
Theophylline	10 mg/dL	0.56 mmol/L
Acetaminophen	20 mg/dL	1.32 mmol/L

* These substances are intended to be representative

f. Assay cut-off:
Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

The method comparison with the predicate device followed NCCLS Guideline EP9-A2 Method Comparison and Bias Estimation Using Patient Samples. A total of 110 human whole blood samples were assayed using the VITROS Chemistry Products %A1c assay and the A1c 2.2 Plus Glycohemoglobin Assay. Each sample was measured in duplicate with two lots of reagent on the VITROS 5, 1 FS Chemistry System. Single replicates of each sample were analyzed on the Tosoh A1c 2.2 Plus Glycohemoglobin Analyzer.

The relationship between the two methods was determined by least squares linear regression with patient sample concentrations for the VITROS %A1c assay as the “y” variable and those from the comparative method as the “x” variable. All data analyzed was within the reportable range of both the VITROS %A1c assay and Tosoh A1c 2.2 Plus Glycohemoglobin Assay.

NGSP results are derived from the International Federation of Clinical Chemistry (IFCC) %A1c units to NGSP %A1c units using the Master Equation⁵: $NGSP = (0.915 \times IFCC) + 2.15$

The relationship between the two methods, determined by least squares linear regression, was:

VITROS % A1c assay = $1.0374 X - 0.3426$ % A1c (NGSP)
with a correlation coefficient of 0.9872

VITROS % A1c assay = $1.0374 X - 0.2866$ % A1c (IFCC)
with a correlation coefficient of 0.9872

The VITROS %A1c assay has met the requirements of the National Glycohemoglobin Standardization Program and is traceable to the Diabetes Control and Complications Trial Reference Method.

b. Matrix comparison:

Anti-coagulants were evaluated by paired-difference testing. Blood was collected from 5 individuals in tubes containing anti-coagulants. All specimens were stored at room temperature for less than 2 hours and then tested in triplicate from a single cup.

The data was tabulated and reviewed for outliers as per NCCLS Ep5-A⁶. No outliers were identified. The bias between the mean value (n=3) for each test condition (sample type) was calculated using the equation below.

Bias = Test Condition Prediction – Fresh Whole blood with EDTA Prediction

Whole blood with EDTA was used as the control sample because it is the specimen matrix used to establish overall accuracy of the method. Anti-coagulants were deemed acceptable if the bias was within predetermined acceptance criteria.

Sample ID	Anticoagulant Type	Mean (n = 3) %A1c NGSP	SD	Bias to EDTA %A1c NGSP
1	EDTA	12.32	0.10	0.00
2		5.10	0.04	0.00
3		6.16	0.05	0.00
4		6.79	0.02	0.00
5		5.56	0.01	0.00
1	Li-Heparin	12.47	0.10	0.15
2		5.05	0.03	-0.05
3		6.17	0.03	0.01
4		6.75	0.02	-0.04

5		5.60	0.06	0.04
1	Sodium Fluoride/ Potassium Oxalate	12.45	0.03	0.13
2		5.12	0.02	0.02
3		6.19	0.02	0.03
4		6.81	0.06	0.02
5		5.64	0.07	0.08

The bias values for matched patient whole blood samples collected in tubes containing the anticoagulants EDTA, Lithium Heparin and Sodium Fluoride/ Potassium Oxalate were within the predetermined acceptance criteria for the VITROS Chemistry Products %A1c assay.

3. Clinical studies:

a. *Clinical sensitivity:*

Clinical studies are not typically submitted for this device type.

b. *Clinical specificity:*

Clinical studies are not typically submitted for this device type.

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

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

Literature searches were performed to determine published reference intervals for glycated hemoglobin assay methods that are traceable to the National Glycohemoglobin Standardization Program. The reference interval for the VITROS Chemistry Products %A1c Reagent assay is based on the following:

- The VITROS %A1c calibrators' values are traceable to the International Federation of Clinical Chemistry (IFCC) values.
- IFCC and NGSP %A1c values are related by Master Equation:
 $NGSP = (0.915 \times IFCC) + 2.15$.
- NGSP has established %A1c reference intervals, its interpretation, and relationship to mean plasma glucose.
- The NGSP reference intervals are based on the position statement of the American Diabetes Association regarding glycated hemoglobin testing.
- The American Diabetes Association Position Statement on test for glycemia is based on the outcome of the Diabetes Control and Complications Trial Research Group (DCCT). This reference also described the reference population

Reference Interval for VITROS %A1c

%A1c (NGSP)*	Interpretation
<6.0	Non-Diabetic Range
6.0 to 7.0	ADA Therapeutic Target
>7.0	Action Suggested

*NGSP reference intervals were published by the National Glycohemoglobin Standardization Program (NGSP). The intervals are based on the American Diabetes Association (ADA) position statement regarding glycated hemoglobin testing. The reference intervals are applicable to methods that are traceable to the Diabetes Control and Complications Trial (DCCT).

To provide reference range guidance for laboratories reporting results in IFCC units, the following values are calculated for the NGSP reference values using the Master Equation: $NGSP = (0.915 \times IFCC) + 2.15$.

Calculated %A1c (IFCC)	Interpretation
<4.2	Non-Diabetic Range
4.2 to 5.3	ADA Therapeutic Target
>5.3	Action Suggested

Description of Reference Population:

The Diabetes Control and Complications Trial Research Group: The effect of intensive treatment of diabetics on the development and progression of long-term complications in insulin-dependent diabetes mellitus. N Engl J Med 329:977-86; 1993.

N. Conclusion:

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