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

A. 510(k) Number:

k110934

B. Purpose for Submission:

New Device

C. Measurand:

Whole blood hemoglobin A1c (HbA1c)

D. Type of Test:

Quantitative, latex agglutination inhibition method.

E. Applicant:

Siemens

F. Proprietary and Established Names:

ADVIA® 1800 Chemistry Systems Hemoglobin A1c 3 Automated Pretreatment Assay

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
LCP	Class II	21 CFR 864.7470	Hematology (81)

H. Intended Use:

1. Intended use(s):

   See indications for use below.

2. Indication(s) for use:

The ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment Assay is for In Vitro diagnostic use in the quantitative determination of Hemoglobin A1c, a diabetes marker,

in whole blood on the ADVIA Chemistry systems. Such measurements are used for monitoring the long-term care of persons with diabetes. The A1c_3 and total hemoglobin tHb_3 values generated as part of the ADVIA Chemistry HbA1c% and HbA1c R assays are intended for use in the calculation of the A1c/total hemoglobin ratio, and must not be used individually for diagnostic purposes.

3. Special conditions for use statement(s):

   For prescription use only

4. Special instrument requirements:

   Siemens ADVIA 1800 analyzer.

I. Device Description:

The concentration of A1c and the concentration of total hemoglobin are measured and their ratio is reported. The automated pretreatment assays (AVIA 1800 Chemistry A1c_3 and ADVIA 1800 Chemistry tHb_3) use 3 ADVIA Chemistry reagents:

- A1c_3 Agglutinator/Total Hemoglobin reagent (A1c_3 R1)
- A1c_3 Antibody Reagent (A1c_3 R2)
- A1c_3 Denaturant Reagent (A1c_3 DENAT)

A1c_3 R1 reagent contains A1c hapten covalently attached polymer, bovine serum albumin, buffer, sodium azide as a preservative and surfactant.

Alc_3 R2 reagent contains Anti-HbA1c antibody coupled with latex (mouse, monoclonal), bovine serum albumin, buffer, surfactant and preservative (5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one)

A1c_3 DENAT reagent contains Porcine pepsin, buffer and preservative (5-chlor-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one)

J. Substantial Equivalence Information:

1. Predicate device name(s):

   ADVIA® Chemistry Hemoglobin A1c Assay

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

   k081895

3. Comparison with predicate:

Similarities and Differences		
Item	New Device ADVIA® 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment Assay	Predicate ADVIA® Chemistry Hemoglobin Assay k081895
Intended Use	For *in vitro* diagnostic use in the quantitative determination of Hemoglobin A1c, a diabetes marker, in whole blood on the ADVIA Chemistry systems. Such measurements are used for monitoring the long-term care of persons with diabetes	Same
Test Principle	**Hemoglobin:** measurement of released heme in the Soret region at 410 nm **HbA1c:** Latex agglutination inhibition	**Hemoglobin:** Conversion of all hemoglobin derivatives into alkaline hematin. **HbA1c:** Latex agglutination inhibition
Expected Values	4-6% (20-42 mmol/mol)	Same
Format	Liquid, ready for use	Same
Sample type	Human whole blood (lithium heparin or potassium EDTA)	Same
Calibrators	Siemens ADVIA® Chemistry A1c Calibrators	Same
Measurement Wavelength	A1c_3: 694 nm tHb_3: 410/694 nm	A1c: 694 nm tHb_2: 596 nm
Analytical range	2.9- 15.4% (8-144 mmol/mol)	Same

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

CLSI EP5-A2: Evaluation of Precision Performance of Clinical Chemistry Devices

CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation

L. Test Principle:

The ADVIA® 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment Assay is a latex agglutination inhibition assay that is used for the measurement of specific A1c.

In an automated pretreatment step, the whole blood sample is mixed with the A1c_3 Denaturant Reagent. The red blood cells are lysed and the hemoglobin chain is hydrolyzed

by the protease present in the reagent. For the measurement of total hemoglobin, the A1c_3 Agglutinator Reagent (A1c_3 R1) is used. The assay is based on the determination of released heme in the Soret region at 410 nm.

A latex agglutination inhibition assay is used for the measurement of specific A1c. A second protease in the R1 reagent further hydrolyzes the HbA1c sample to a glycated pentapeptide, which competes with the agglutinator (synthetic polymer containing multiple copies of the immunoreactive portion of A1c) for the anti-HbA1c antibody, thereby reducing the rate of agglutination. A concentration curve is obtained by monitoring the change the scattered light at 694 nm as a change of absorbance. The actual change in absorbance is inversely proportional to the concentration of A1c in the sample.

The A1c_3 and thB_3 results use the HbA1c% ratio to determine HbA1c results in NGSP equivalent units (%). The HbA1cR Ratio is used to determine HbA1c results in IFCC equivalent units (mmol/mol). The required assays and parameter sheets are summarized in the following table:

Result Units	**Photometric Assays**	**Ratio Assays**
NGSP (%)	A1c_3 and tHb_3 HbA1c%	HbA1c%
IFCC (mmol/mol)	A1c_3 and tHb_3	HbA1cR

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   a. Precision/Reproducibility:

   A precision study was performed by testing a normal blood pool, high blood pool, two whole blood pools (6.5% and 10% HbA1c) and two levels of control material. Each sample was assayed 2 replicates per run, 2 runs per day, for at least 20 days on the ADVIA 1800 analyzer. The results are shown below:

Sample	N	Mean (%A1c)	Within Run		Total Imprecision	
			SD	%CV	SD	%CV
Control 1	80	5.45	0.07	1.3	0.13	2.4
Control 2	80	8.70	0.06	0.7	0.14	1.6
Normal Blood Pool	80	5.07	0.07	1.3	0.12	2.4
High Blood Pool	80	7.52	0.06	0.8	0.14	1.8
Whole Blood Pool (6.5%)	80	6.19	0.04	0.7	0.12	1.9
Whole Blood Pool (10%)	80	10.61	0.09	0.8	0.14	1.3

An additional precision study was performed in order to show the precision of the

assay at the higher limits of the assay range. Two human whole blood samples were assayed. Each sample was assayed 2 replicates per run, 2 runs per day for 7 days on the ADVIA 1800 analyzer. The results are shown below:

Sample	N	Mean (%A1c)	Within Run		Total Imprecision	
			SD	%CV	SD	%CV
Diabetes Whole Blood Control 1	28	12.1	0.07	0.6	0.11	0.9
Diabetes Whole Blood Control 2	28	14.8	0.08	0.5	0.17	1.1

b. *Linearity/assay reportable range:*

A commercially available linearity set solution was used to assess the linearity across the entire measuring range of the assay. The low and high levels of the linearity set were mixed to make a total of nine intermediate levels on the ADVIA 1800 analyzer using the Hemoglobin A1c3 Automated Pretreatment Assay. The range of samples tested was 1.13 – 19.44 %A1c. The observed values versus the expected values are 95.0 – 100.7%. The regression was:

y= 1.0487x – 0.2829, r=0.999.

The data provided support the sponsor's claims that the reportable range of this assay is 2.9 – 15.4% A1c.

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

The ADVIA 1800 Chemistry Hemoglobin A1c_3 assay standardization is traceable to the IFCC reference calibrators. The assigned A1c and total hemoglobin (tHb_3) values of the calibrators are traceable to the NGSP reference method (Tosoh G7) through a correlation study. The ADVIA Chemistry Hemoglobin A1c assay is certified with the National Glycohemoglobin Standardization Program (NGSP). The NGSP certification expires in one year. See NGSP website for current certification at http://www,ngsp.org.

The derived result of the ratio (%) from the NGSP correlation is calculated from the individual quantitative results for total hemoglobin (tHb_3) and Hemoglobin A1c (HbA1c_3). The International Federation of Clinical Chemistry (IFCC) units of mmol/mol are calculated using the Master Equation:

IFCC= (NGSP-2.15)/0.092

Two different units are provided to the customers:

NGSP equivalent units (%) and IFCC equivalent units (mmol/mol)

Stability:

Shelf-life and open vial stability testing protocols for the calibrators have been previously cleared under k081895.

d. Detection limit:

The limit of Blank (LoB) and Limit of Detection (LoD) were determined by assaying a zero sample (blank) and five low HbA1c samples according to CLSI guideline EP17-A. Each sample was assayed twice a day in replicates of ten (n=60) for three days on the ADVIA analyzer. The detection limits are summarized in the table below.

Platform/Method	LoB (%A1c)	LoD (%A1c)
ADVIA® 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment Assay	0.77	0.91

The assay has a reportable range of 2.9% to 15.4% A1c.

e. Analytical specificity:

i.) Studies were performed to assess common or known substances that could interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay. The interfering substances were evaluated in whole blood EDTA samples that had different Hgb A1c concentrations. The low range samples ranged from 4.87 – 5.26%HbA1c (30-34 mmol/mol); the high level samples ranged from 8.99 – 9.90% HbA1c (74-84 mmol/mol). Samples were assayed in duplicate on the ADVIA 1800 analyzer using the Hemoglobin A1c_3 assay. The sponsor's acceptance criterion is analyte recovery should not vary from the base recovery by more than 10%. No significant interference was defined as the % recovery of <10%.

The sponsor claimed that there was no significant interference by the following interferents:

- Conjugated and unconjugated bilirubin up to 60 mg/dL
- Triglycerides (Intralipid) up to 1000 mg/dL
- Rheumatoid Factor up to 936 IU/mL

ii.) An interference study was performed to assess the affect of labile A1c with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay. Two levels of pooled blood samples (~5.3 and 9.9% A1c) were used and each pool was split into two aliquots. One aliquot was used as the control sample while the other aliquot was supplemented by dissolving 0.030 g (30mg) of glucose into the pool sample (glucose concentration = 1500mg/dL). The sample was incubated for five hours at 37˚C to facilitate formation of labile A1c. Both aliquots were tested on the ADVIA 1800 analyzer. The sponsor's acceptance criteria is ≤ 10% bias

between the tested and the control samples.

The sponsor concluded that labile A1c concentrations do not interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay.

iii.) An interference study was performed to assess the effect of urea with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay. Two levels of pooled whole blood samples (~5.1 and 9.7 A1c) were used and each pool was split into two aliquots. One aliquot was used as the control sample while the other was supplemented by dissolving 3 mg of urea into the pool sample (urea concentration = 150 mg/dL). The sample was incubated for two hours at 37˚C. Both aliquots were tested on the ADVIA 180 analyzer. The sponsor's acceptance criterion is ≤ 10% bias between the tested and the control samples.

The sponsor concluded that urea concentrations ≤ 150 mg/dL do not interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay.

iv.) An interference study was performed to assess the effect of acetylsalicylic acid with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay. One pooled whole blood sample (~5.9% A1c) was used and split into two aliquots. One aliquot was used as the control sample while the other aliquot was supplemented by dissolving 900.8 mg of acetylsalicylic acid into an ethanol stock solution and then adding this stock solution into the pool sample. The sample was incubated for three hours at 37˚C. Both aliquots were tested on the ADVIA 1800 analyzer. The sponsor's acceptance criterion is ≤ 10% bias between the tested and the control samples.

The sponsor concluded that acetylacetic acid concentrations ≤ 10mM does not interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay.

v.) An interference study was performed to assess the effect of carbamylated A1c with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay. A pooled whole blood sample (~4.78 %A1c) was split into two aliquots. One aliquot was used as the control sample while the other aliquot was supplemented by dissolving 325 mg of sodium cyanate into H_2O to make a stock solution in which 2.5 ml of this stock solution is then added to the whole blood sample. The samples were incubated for one hour at 37˚C for one hour. Both aliquots were tested in duplicate on the ADVIA 1800 analyzer. The sponsor's acceptance criterion is ≤ 10% bias between the tested and the control samples.

The sponsor concluded that carbamylated A1c does not interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay.

vi.) A hemoglobin concentration interference study was done with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay by testing varying

concentrations of total hemoglobin at the same level of %A1c. Seven individual normal donors were used for the study. The acceptance criterion for the hemoglobin interference was %HbA1c at different hemoglobin levels must be within **±7.5%** of the target % HbA1c. The results met the sponsor's predetermined criteria and the sponsor concluded that total hemoglobin in the range of 7 to 26 g/dL does not interfere with the ADVIA 1800 Chemistry Hemoglobin A1c_3 Automated Pretreatment assay.

vii.) A hemoglobin variant interference study was carried out using NGSP samples known to contain Hemoglobin variants C, S, D, E and F. These variant samples were tested on the Primus Ultra 2 HPLC system and the ADVIA 1800 analyzer with the ADVIA Hemoglobin A1c_3 Automated Pretreatment method. All the variants tested showed <10% bias at 6% and 9% except Hemoglobin F at concentrations greater than 10%. Hemoglobin F variant >10% showed a negative bias therefore, the sponsor has the following limitation in their labeling:

"Fetal Hemoglobin (HbF) consists of two alpha and two gamma chains that are not recognized by the anti-HbA1c antibody. Samples that contain high amounts of HBF (>10%), usually found in some people with thalassemia, in infants, and in some pregnant women, may yield a lower than expected HbA1c result with this assay. For blood samples containing HbF (>10%), HbA1c results obtained by this assay should not be compared to published normal or abnormal results".

See NGSP at http://www.ngsp.org for assay interferences with hemoglobin variants.

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study was performed in conjunction with the NGSP guidelines for manufacturer certification using the reference laboratory. A method comparison study was performed versus the ADVIA Chemistry HbA1c method (k081895). The study was completed using 98 whole blood EDTA samples which were obtained by a commercial vendor and value assigned by the NGSP. Each sample was analyzed in duplicate on the ADVIA 1800 using the candidate device and the predicate device. Sample range tested was 3.14 – 14.92% A1c. The linear regression correlation using only the first set of replicates is as follows:

System	n	Regression Equation	r	Sy.x
ADVIA 1800	98	y=1.00x-0.27	0.994	0.37

An additional separate study was done versus the Tosoh G7 Automated HPLC analyzer, a secondary reference method for the NGSP. The second study was performed using 68 whole blood EDTA samples which were obtained from a commercial vendor and value assigned by the NGSP. Each sample was analyzed in duplicate. The linear regression correlation is as follows:

System	n	Regression Equation	r	Sy.x	Sample Range
ADVIA 1800	68	y=0.9682x+0.2349	0.997	0.16	4.5-12.5

(x=NGSP Reference Method (Tosoh G7), y=ADVIA 1800

b. Matrix comparison:

A total of 45 random matched sample pairs (Potassium EDTA, Lithium heparin) were tested on the ADVIA 1800 using the Automated Pretreatment HbA1c_3 method. The linear regression correlation is presented in the table below:

System	Regression Equation	r	Sy.x	Sample Range (%)
ADVIA 1800	y=1.01x-0.03	0.999	0.09	2.9-14.15

(x=Potassium EDTA, y=Lithium heparin)

The sponsor recommends that whole blood preserved with potassium EDTA and Lithium heparin be used with this assay.

3. Clinical studies:

a. Clinical Sensitivity:

Not applicable

b. Clinical specificity:

Not applicable

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

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

   The expected normal %HbA1c range is 4.0-6.0% (20-92 mmol/mol)* according to the literature cited.

   *Wu AHB. *Tietz Clinical Guide to Laboratory Tests, 4th edition*, Saunders Elsevier, St. Louis, MO 480-483 (2006)

N. Proposed Labeling:

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

O. Conclusion:

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