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

A. 510(k) Number:

K160724

B. Purpose for Submission:

New Device

C. Measurand:

Creatine Kinase

D. Type of Test:

Quantitative photometric enzyme assay

E. Applicant:

Siemens Healthcare Diagnostics, Inc.

F. Proprietary and Established Names:

ADVIA Chemistry® Creatine Kinase (CK_L) Assay

ADVIA Chemistry® Enzyme 3 Calibrator

G. Regulatory Information:

1. Regulation section:

   21 CFR §862.1215 - Creatine phosphokinase/creatine kinase or isoenzymes test system

   21 CFR §862.1150 – Calibrator, Secondary

2. Classification:

   Class II

3. Product code:

   CGS, JIT

4. Panel:

   Clinical Chemistry

H. Intended Use:

1. Intended use(s):

   See indications for use statement below.

2. Indication(s) for use:

   The ADVIA Chemistry® Creatine Kinase assay is for in vitro diagnostic use in the quantitative determination of creatine kinase activity in human plasma (lithium heparin)

or serum on ADVIA Chemistry systems. The assay can be used to aid in the diagnosis and treatment of myocardial infarction and muscle diseases, such as Duchenne progressive muscular dystrophy.

ADVIA Chemistry® Enzyme 3 Calibrator is intended for in vitro diagnostic use in the calibration of the ADVIA Chemistry Creatine Kinase (CK_L) assay on the ADVIA Chemistry systems.

3. Special conditions for use statement(s):

For in vitro diagnostic and prescription use only.

4. Special instrument requirements:

ADVIA Chemistry 1800 System.

I. Device Description:

ADVIA Chemistry Creatine Kinase (CK_L) assay is a ready-to-use liquid reagent packaged for use on the automated ADVIA Chemistry systems. The reagent kit consists of reagent 1 and reagent 2.

Reagent 1 contains, 0.09% sodium azide, imidazole buffer 123mM, pH 6.5, 2.46mM EDTA, 2.46mM ADP, 6.14mM AMP, 19µM diadenosine pentaphosphate, 2.46mM NADP, ≥4U/mL hexokinase, ≥2.8U/mL G-6-PDH, 24.6mM N-acetyl-L-cysteine and 12.3mM Mg^{2+}.

Reagent 2 contains, buffer 20mM, pH 8.8, 120mM glucose, 184mM creatine phosphate, 2.46mM EDTA and 0.09% sodium azide.

ENZ 3 CAL is a liquid frozen human serum albumin based product containing creatine kinase MM from human heart. Enzyme 3 Calibrator kit consists of six vials of the same calibrator, 2mL per vial, and is ready for use.

J. Substantial Equivalence Information:

1. Predicate device name(s):

ADVIA Chemistry Creatine Kinase (CKNAC) Assay

Dimension Vista ENZ 6 CAL

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

K991576

K083579

3. Comparison with predicate:

Item	ADVIA Chemistry Creatine Kinase (CK_L) Assay (Candidate Device)	ADVIA Chemistry Creatine Kinase (CKNAC) Assay (K991576) (Predicate Device)
Intended Use	For in vitro diagnostic use in the quantitative determination of creatine kinase activity in human plasma (lithium heparin)	Same

Item	ADVIA Chemistry Creatine Kinase (CK_L) Assay (Candidate Device)	ADVIA Chemistry Creatine Kinase (CKNAC) Assay (K991576) (Predicate Device)
	or serum on ADVIA Chemistry systems.	
Assay principle	Creatine Kinase reacts with creatine phosphate and ADP to form ATP which is coupled to the hexokinase-G6PD reaction, generating NADPH. The concentration of NADPH is measured by the increase in absorbance at 340/596 nm.	Creatine Kinase reacts with creatine phosphate and ADP to form ATP which is coupled to the hexokinase-G6PD reaction, generating NADPH. The concentration of NADPH is measured by the increase in absorbance at 340/410 nm.
Kit Components	Reagent 1, liquid Reagent 2, liquid	Reagent 1, lyophilized Reagent 1 mix, lyophilized
Calibrator	ADVIA Enzyme 3 Calibrator	None, fixed calibrator value
Sample Type	Serum and lithium heparinized plasma	Same
Analytical Range	15 to 1300 U/L	0 to 1300 U/L
Extended Range	1300 to 7800 U/L	Same
Analyzers	ADVIA Chemistry 1800 Systems	ADVIA Chemistry 1650, 1200, 1800, 2400 and XPT Systems

Item	ADVIA Chemistry® ENZ 3 CAL (Candidate Device)	Dimension Vista® ENZ 6 CAL (K083579) (Predicate Device)
Intended Use	For *in vitro* diagnostic use for the calibration of ADVIA Chemistry Creatine Kinase (CK_L) assay on the ADVIA Chemistry® systems.	For *in vitro* diagnostic use for the calibration of Creatine Kinase (CKI) and Creatine Kinase MB (MBI) methods on the Dimension Vista® System.
Traceability	To the IFCC Reference Method	Same
Composition	ADVIA Chemistry ENZ 3 CAL is a liquid frozen human serum albumin (5%) and preservatives with lot-specific concentrations of creatine kinase MM from human heart.	ENZ 6 CAL is a liquid frozen human serum albumin based product containing creatinine kinase BB from porcine brain and creatine kinase MM from human heart.

Item	ADVIA Chemistry® ENZ 3 CAL (Candidate Device)	Dimension Vista® ENZ 6 CAL (K083579) (Predicate Device)
Target Concentration	650 U/L	1050 U/L
Package Content	6 vials of calibrator, 2mL per vial	3 vials: Calibrator A, 2mL per vial
Storage	-25 to -15°C	-20 °C or below

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

Clinical and Laboratory Standards Institute (CLSI) EP09-A3, Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline, 3rd Edition.

CLSI EP05-A2, Evaluation of Precision Performance of Quantitative Measurement Methods: Approved Guideline, 2nd Edition.

CLSI EP06-A, Evaluation of Linearity of Quantitative Measurement Procedures: a Statistical approach, 1st Edition.

CLSI EP17-A2, Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures, 2nd Edition.

CLSI EP07-A2, Approved Guideline Interference Testing in Clinical Chemistry, 2nd Edition.

CLSI EP28-A3c, Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory, 3rd Edition.

CLSI EP25-A, Evaluation of Stability of In Vitro Diagnostic Reagents; Approved, 1st Edition.

L. Test Principle:

Creatine kinase reacts with creatine phosphate and ADP to form adenosine triphosphate (ATP), which is coupled to the hexokinase-G6PD reaction, generating NADPH. The concentration of NADPH is measured by the increase in absorbance at 340/596 nm.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   All analytical performance studies presented below were performed on the ADVIA Chemistry 1800 system. All studies were performed using serum samples collected in serum separator tubes (SST).

   a. *Precision/Reproducibility:*

   Precision testing was performed according to the CLSI EP05-A2 guidelines. Samples consisted of three commercial quality controls, three patient serum pools and one patient plasma pool. Testing was performed over twenty days, two runs per day, two tests per run, using three lots of the ADVIA Chemistry CK_L assay on the ADVIA Chemistry 1800 system (N=80 per lot). The precision study results are summarized

below.

Lot 1:

Sample	Mean (U/L)	Repeatability		Total	
		SD	%CV	SD	%CV
QC1	75	1.3	1.7	1.9	2.6
QC2	232	1.5	0.6	3.1	1.4
QC3	639	2.6	0.4	8.1	1.3
Plasma Pool	1202	4.7	0.4	6.3	0.5
Serum Pool 1	85	1.7	2.0	2.9	3.4
Serum Pool 2	194	3.0	1.5	3.2	1.7
Serum Pool 3	938	5.2	0.6	5.6	0.6

Lot 2:

Sample	Mean (U/L)	Repeatability		Total	
		SD	%CV	SD	%CV
QC1	75	1.0	1.3	1.9	2.5
QC2	233	1.5	0.7	3.4	1.5
QC3	641	3.3	0.5	7.8	1.2
Plasma Pool	1203	4.0	0.3	6.2	0.5
Serum Pool 1	85	1.8	2.1	2.9	3.4
Serum Pool 2	194	2.8	1.5	3.2	1.7
Serum Pool 3	939	5.8	0.6	5.9	0.6

Lot 3:

Sample	Mean (U/L)	Repeatability		Total	
		SD	%CV	SD	%CV
QC1	76	1.3	1.7	1.9	2.5
QC2	235	1.8	0.8	2.9	1.2
QC3	644	3.0	0.5	8.2	1.3
Plasma Pool	1212	4.8	0.4	7.0	0.6
Serum Pool 1	88	1.9	2.2	3.7	4.3
Serum Pool 2	196	2.6	1.3	3.1	1.6
Serum Pool 3	945	5.3	0.6	5.7	0.6

b. *Linearity/assay reportable range:*

A linearity study was conducted following the CLSI EP06-A guidelines. A set nine samples ranging from 1 to 1372 U/L were prepared by serial dilution of a high concentration sample of creatine kinase using a low concentration sample. Each dilution was assayed in replicates of three. Data were analyzed using weighted linear regression analysis and a 2nd and 3rd order polynomial regressions of the mean observed analyte values vs. expected concentrations were generated. The result of regression analysis is shown below.

Regression equation, $y = 1.00x + 0.5$, $R = 1.00$

The linearity study data supports the claimed measuring range of 15 to 1300U/L.

Dilution Recovery Studies: Studies were performed to analyze the recovery of samples containing high levels of creatine kinase outside the claimed measuring range of 15 to 1300U/L by diluting the sample 6-fold and retesting with the candidate device. The high level sample dilution can be performed either manually or using the automated dilution feature on the ADVIA Chemistry 1800 system.

Automated Dilution Recovery – Five serum samples with creatine kinase values greater than the ADVIA Chemistry CK_L assay range of 15 to 1300U/L were assayed with ADVIA Chemistry CK_L assay on the ADVIA Chemistry 1800 system using the automated dilution feature. In addition, the same creatine kinase sample were manually diluted with saline at an equivalent dilution ratio (1:6) as the automated feature, and assayed on with the ADVIA Chemistry CK_L assay on the ADVIA Chemistry 1800 system. The values obtained using the ADVIA Chemistry CK_L automated dilution feature were then compared to the values that were obtained from the manual dilution. A summary of the automated dilution data is presented below.

Test Sample	Automated Dilution with Saline (U/L)	1:6 Manual Dilution with Saline (U/L)	Percent Recovery (Automated vs Manual) ADVIA CK_L
1	2314	2280	102
2	2265	2270	100
3	1350	1322	102
4	3633	3540	103
5	2948	2890	102

Manual Dilution Recovery – Five serum samples with creatine kinase values greater than the ADVIA Chemistry CK_L assay range of 15 to 1300U/L were diluted manually at a 1:6 dilution with saline and assayed on the ADVIA Chemistry 1800 system. In addition, the creatine kinase concentrations were determined using the automated dilution function of the cleared ADVIA CKNAC assay on the ADVIA Chemistry 1800 system. The values obtained using the manual ADVIA Chemistry CK_L assay dilution were then compared to the values that are obtained from the

automated ADVIA CKNAC dilution. A summary of the manual dilution data is presented below.

Test Sample	Automated Dilution with Saline (U/L) CKNAC	1:6 Manual Dilution with Saline (U/L) CK_L	Percent Difference CK_L Manual vs CKNAC Automated
1	2313	2286	99
2	2274	2274	100
3	1343	1356	101
4	3602	3540	98
5	2949	2934	99

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

Assay Traceability - The ADVIA Chemistry CK_L assay is traceable to the SI units (mkat/L) through the IFCC Primary Reference Method for CK via patient sample correlation.

Calibrator Traceability - The assigned values of the ADVIA Chemistry ENZ 3 Calibrator is traceable to the SI units through the IFCC Primary Reference Method for Creatine Kinase. Traceability chain is established using the primary IFCC reference procedure and primary reference material of endogenous patient sample anchor pools.

Calibrator Value Assignment - The ADVIA ENZ 3 calibrator base material is a 5% human serum albumin (HSA) solution with preservatives. An enzyme stock solution, used to spike the calibrators, is prepared in HSA matrix. This stock solution is gravimetrically spiked at a target concentration of creatine kinase MM isoform from human heart. The stock solution concentration is determined by comparing the recovery of the stock solution versus the Master Lot assigned bottle values. Calculated quantities of the stock solution are added to the base to target concentration to produce the calibrator lot. The concentration of CK is verified to be within acceptable range by using an instrument calibrated with Master Lot Calibrators.

Stability of calibrators - The ADVIA ENZ 3 calibrator shelf life and open vial stability studies are performed following the CLSI EP25-A guidelines. The predetermined acceptance criteria and protocols were reviewed and found to be acceptable. The ADVIA ENZ 3 calibrator has a target shelf life of 12 months when stored at -25 to -20ºC. The calibrator must be thawed at room temperature before use. Once the cap is removed, the assigned values are stable for 30 days when recapped immediately and stored at 2-8 ºC. The sponsor states that the ongoing stability monitoring will be performed to assess the stability of the ADVIA ENZ 3 calibrator using real time data at each time point tested throughout the claimed shelf life of the product. The acceptance criteria were reviewed and found to be acceptable..

d. *Detection limit:*

A limit of blank (LoB) study was conducted using five blank samples according to CLSI EP17-A2. Deionized water is used as the blank sample. The samples were tested in replicates of five on the ADVIA Chemistry 1800 system over three days using three lots (N=75 per lot). LoB was calculated non-parametrically. Based on the results, the LoB claim was 3 U/L.

A limit of detection study (LoD) was conducted using five low level samples (serum samples diluted with saline). These samples were prepared by diluting five separate pools of serum with saline to obtain five samples at 5, 6, 7, 8 and 9 U/L of creatine kinase. These samples were tested in replicates of five on the ADVIA Chemistry 1800 system over three days using three lots (N=75 per lot). LoD was calculated parametrically following the CLSI guidelines. Based on the results, the LoD claim was 6 U/L.

A limit of quantitation study (LoQ) was conducted using the ADVIA CK_L assay on the ADVIA Chemistry 1800 system according to CLSI EP17-A2. Four separate serum samples were prepared to target the desired LoQ at approximately 15 U/L and frozen. Each samples was tested once per day in replicates of five using three reagent lots over three days (N=15 per lot). The mean value, standard deviation (SD), bias from the assigned value (on the IFCC traceable ADVIA Creatine Kinase assay CKNAC) and total error (TE) was calculated for each sample across all replicates for each reagent lot. The TE is calculated using the formula TE = 2SD + Bias. The LoQ for the method was taken as the concentration value that gave the highest TE obtained for each reagent lot within a defined total error. The LoQ for the ADVIA Chemistry CK_L assay is 15 U/L.

e. *Analytical specificity:*

Interference testing was performed according to CLSI EP07 to determine the effects of various endogenous and exogenous substances on the ADVIA Chemistry CK_L assay using the ADVIA Chemistry 1800 system. For all interferents, the percent bias was determined by testing a control sample without the interferent and comparing it to the test sample spiked with the interferent. Each sample was tested in triplicate. Interferents were tested at two levels of creatine kinase, 95±14 U/L and 265±40 U/L. For each spiked sample, the percent recovery was determined using the formula,

% Recovery = [(Test result – control result) / control result] X 100

A recovery of ≤10% of control value was defined as non-significant interference. The results of the highest concentration tested without significant interference are summarized in the table below.

Interferent	Interferent Test Concentration	Creatine Kinase Test Concentration (U/L)	Percent Difference
Hemolysate (hemoglobin)	125 mg/dL	100	7%
	125 mg/dL	284	1%
Bilirubin Conjugated	60 mg/dL	93	1%
	60 mg/dL	256	2%
Bilirubin Unonjugated	60 mg/dL	94	2%
	60 mg/dL	264	2%
Lipemia (Intralipid)	1000 mg/dL	94	5%
	1000 mg/dL	276	0%
Ascorbic Acid	6 mg/dL	91	4%
	6 mg/dL	271	1%
Sulfasalazine	30 mg/dL	94	0%
	30 mg/dL	264	2%
Sulfapyridine	30 mg/dL	93	1%
	30 mg/dL	263	2%

   f. *Assay cut-off:*

     Not applicable.

2. Comparison studies:

   a. *Method comparison with predicate device:*

     A split sample method comparison between the ADVIA Chemistry CK_L assay versus the predicate ADVIA Chemistry CKNAC assay on the ADVIA Chemistry 1800 system was performed following the CLSI EP09-A3 guidelines. A total of 116 native human serum samples were assayed in singlicate across the assay range of 15 to 1300U/L. A total of ten samples were altered of which eight samples were diluted with saline and two samples were altered by mixing two samples. None of the samples were spiked samples. Analysis of the results using Deming regression yielded the following:

     Y= 1.01x - 1.8, R = 1.00.

     Slope (95% CI) = 1.01 (1.00 to 1.02)

     Intercept U/L (95% CI) = -1.8 (-3.3 to -0.3)

     X= ADVIA 1800 - CKNAC (predicate device); Sample range, 23 to 1253 U/L.

     Y= ADVIA 1800 – CK_L (candidate device); Sample range, 22 to 1280 U/L.

Method Comparison versus IFCC Reference Method – A split sample method comparison between the ADVIA Chemistry CK_L assay on the ADVIA Chemistry 1800 system versus the IFCC certified creatine kinase reference method on the Konelab 30i analyzer (Thermo Fisher Scientific) was performed following the CLSI EP09-A3 guidelines. One hundred remnants de-identified human serum samples were assayed in duplicate across the assay measuring range of 15 to 1300U/L; however, only the first result was used in each analysis. Ten samples were diluted with saline and no samples were spiked. The results were analyzed by Deming (Orthogonal) regression. A summary of the IFCC method comparison data is presented below.

Y= 1.05x – 6.9, R = 1.00.

Slope (95% CI) = 1.05 (1.03 to 1.07)

Intercept U/L (95% CI) = -6.9 (-8.8 to -4.9)

X= IFCC Reference Method (predicate device); Sample range, 21.00 to 1178.24 U/L.

Y= ADVIA 1800 – CK_L (candidate device); Sample range, 16.00 to 1245 U/L.

b. *Matrix comparison:*

To characterize correlation between lithium heparin plasma and serum samples from the whole blood samples collected in serum separator tubes (SST), a matrix comparison study was performed using paired samples on the ADVIA Chemistry System using the ADVIA Chemistry CK_L assay. Fifty sets of native samples with serum creatine kinase level ranging from 37 to 1282 U/L. Five sets were spiked with creatine kinase stock solution to cover the measuring range up between 859 U/L to 946 U/L. Two samples were excluded; one for hemolysis and the second was outside the assay measuring range. Deming regression analysis was used to fit the ADVIA CK_L results of the Li-heparinized plasma samples versus the SST serum samples. A summary of the expected values is presented below.

Y= 1.01x – 0.6, R = 1.00.

Slope (95% CI) = 1.01 (0.99 to 1.03)

Intercept U/L (95% CI) = -0.6 (-2.5 to 1.3)

X= SST Serum; Sample range, 37 to 1282 U/L.

Y= Li-heparinized Plasma; Sample range, 39 to 1284 U/L.

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:

   A study was performed to evaluate whether the reference ranges cited in Tietz[1] (female reference range: 34-145 U/L and male reference range: 46-171 U/L) could be transferred to the ADVIA Chemistry CK_L assay. The study was performed in accordance with CLSI EP28-A3c (section 11). Based on the study results, both the male and female reference ranges from Tietz were transferred to the ADVIA CK_L assay.

   [1]Burtis CA, Ashwood ER, and Bruns DE. Tietz Textbook of Clinical Chemistry and Molecular Diagnostics. 5th ed. St. Louis, MO: Saunders Elsevier; 2012.

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.