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

A. 510(k) Number:

K112343

B. Purpose for Submission:

To obtain a substantial equivalence determination for the ADVIA Centaur Syphilis Assay

C. Measurand:

Due to an administrative error, the reference to IgG was erroneously included and is now removed.

Antibodies to *Treponema pallidum (T. pallidum)*

D. Type of Test:

Direct sandwich chemiluminometric immunoassay

E. Applicant:

Siemens Healthcare Diagnostics, Inc.

F. Proprietary and Established Names:

ADVIA Centaur Syphilis assay

G. Regulatory Information:

1. Regulation section:

   21 CFR § 866.3830, *Treponema pallidum* treponemal test reagents

2. Classification:

   Class II

3. Product code:

   LIP – Enzyme linked immunoabsorption assay, *Treponema pallidum*

4. Panel:

   Microbiology (83)

H. Intended Use:

1. Intended use:

The ADVIA Centaur Syphilis (SYPH) assay is an *in-vitro* diagnostics immunoassay for the qualitative determination of antibodies to *Treponema pallidum* in human serum or plasma (EDTA, lithium or sodium heparinized, citrate) using the ADVIA Centaur® and ADVIA Centaur® XP systems as an aid in the diagnosis of syphilis.
The ADVIA Centaur Syphilis assay is not intended for blood and tissue donor screening.

ADVIA® Centaur Syphilis Quality Control Materials are for *in-vitro* diagnostic use to monitor the performance of the Syphilis assay on the ADVIA Centaur® systems.
The performance of the SYPH quality control material has not been established with any other Syphilis assay.

2. Indications for use:

The ADVIA Centaur Syphilis (SYPH) assay is an *in-vitro* diagnostics immunoassay for the qualitative determination of antibodies to *Treponema pallidum* in human serum or plasma (EDTA, lithium or sodium heparinized, citrate) using the ADVIA Centaur® and ADVIA Centaur® XP systems as an aid in the diagnosis of syphilis.
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The performance of the SYPH quality control material has not been established with any other Syphilis assay.

3. Special conditions for use statement:

For prescription use only

4. Special instrument requirements:

The ADVIA Centaur and the ADVIA Centaur XP systems

I. Device Description:

The ADVIA Centaur syphilis assay is a fully automated, antigen sandwich assay, using direct chemiluminometric technology. The syphilis kit contains the following:

- 1 ReadyPack® primary reagent pack containing ADVIA Centaur Syphilis Solid Phase Reagent (20 mL);
- 1 Ancillary pack containing ADVIA Centaur Syphilis Ancillary Reagent (10mL)
- ADVIA Centaur Syphilis Master Curve card
- 2 vials of Syphilis Low Calibrator (2 mL fill volume)
- 2 vials of Syphilis High Calibrator (2 mL fill volume)

- ADVIA Centaur Syphilis Calibrator Assigned Value cards

In addition Syphilis quality control materials (2 vials of negative control and 2 vials of positive control with 7 mL fill volume each) are provided separately.

J. Substantial Equivalence Information:

1. Predicate device name:
   Immulite 2000 Syphilis Screen

2. Predicate 510(k) number:
   K091361

3. Comparison with predicate:

   Due to an administrative error, the reference to IgG for device similarities was erroneously included and is now removed.

Similarities		
Item	Device	Predicate
Intended Use	Intended for the qualitative determination of antibodies to *Treponema pallidum*	Same
Assay type	Direct sandwich immunoassay based on chemiluminescent technology	Enzyme labeled, one step chemiluminescent immunoassay
Cut-offs	< 0.9 Non-reactive ≥ 0.9 to < 1.1 Equivocal ≥ 1.1 Reactive	Same
Sample volume	100 µL	Same

Differences		
Item	Device	Predicate
Sample type	Serum, heparinized plasma, EDTA plasma, citrate plasma	Serum, heparinized plasma only
Instrument used	ADVIA Centaur	Immulite 2000
Capture/Detection Antigen/Antibody	Recombinant antigens TpN17 and TpN15 as biotin conjugates and recombinant antigens TpN17 and TpN15 as acridinium ester conjugates	Beads coated with purified recombinant antigen TpN17 are linked to enzyme conjugated purified recombinant TpN17 antigen in the reagent
Calibrators	2 calibrators - liquid in human plasma with a 2ml. fill volume	1 calibrator – lyophilized in human serum with a 4 ml fill volume
Controls	2 liquid controls in human plasma with a 7ml fill volume	2 lyophilized controls in human serum with a 6 ml fill volume

K. Standard/Guidance Document Referenced:

CLSI EP05-A2, Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline.

CLSI GP-10A, Assessment of the Clinical Accuracy of Laboratory Tests using ROC Plots

CLSI EP 12-A2, User Protocol for Evaluation of Qualitative Test

Guidance for Industry and FDA Staff – Assayed and Unassayed Quality Control Material, June 7, 2007.
http://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm
079179.htm

L. Test Principle:

The ADVIA Centaur syphilis assay is a fully automated, antigen sandwich assay, using direct chemiluminometric technology. The ancillary pack reagent containing acridinium-ester-labeled *T. pallidum* recombinant antigens is added to the sample. These *T. pallidum* antigens complex with the antibodies in the sample. The solid phase containing biotinylated T. *pallidum* recombinant antigens preformed to streptavidin-coated magnetic latex particles is then added to the sample. These particles capture the *T. pallidum* antigen-antibody complexes. Antibody-antigen complexes will form if Syphilis antibodies are present in the sample. A direct relationship exists between the level of antibodies to *T. pallidum* present in the patient sample and the amount of relative light units (RLUs) detected by the system. A result of reactive, nonreactive, or equivocal is determined according to the Index Value established with the calibrators.

M. Performance Characteristics:

1. Analytical performance:

a. Precision/Reproducibility:

Precision estimates were computed according to CLSI document EP05-A2, Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline. Within run and total imprecision were evaluated by testing 5 serum based samples (serum sample pools), 3 plasma based samples (2 controls. low and high. and one additional plasma pool) and 2 calibrators (low and high). The elevated levels were spiked with syphilis antigen to achieve appropriate concentrations. The samples were assayed in duplicate over the course of 20 days, two runs per day, for a total of 40 runs and 80 replicates.

Pool	# Repl.	Mean (Index)	Within Run		Between Run		Between Day		Total	
			SD	CV	SD	CV	SD	CV	SD	CV
Calibrator High (plasma) (high positive)	80	8.16	0.14	1.67	0.06	0.78	0.26	3.20	0.30	3.69
Calibrator Low (plasma) (high negative)	80	0.68	0.01	1.81	0.01	0.83	0.02	2.65	0.02	3.32
Control Negative (plasma) (low negative)	80	0.11	0.00	NA	0.00	NA	0.00	NA	0.01	NA
Control Positive (plasma) (moderate positive)	80	3.84	0.06	1.50	0.04	0.99	0.12	3.22	0.14	3.69
Plasma sample (moderate positive)	80	1.99	0.03	1.44	0.02	1.05	0.06	2.93	0.07	3.43
Serum sample 1 (low negative)	80	0.19	0.00	NA	0.00	NA	0.00	NA	0.01	NA
Serum sample 2 (high negative)	80	0.80	0.01	1.16	0.01	0.86	0.02	2.86	0.03	3.20
Serum sample 3 (low positive)	80	1.28	0.02	1.31	0.01	0.93	0.04	2.79	0.04	3.22
Serum sample 4 (high positive)	80	6.96	0.10	1.45	0.07	0.95	0.25	3.63	0.28	4.02
Serum sample 5 (high positive)	80	21.45	0.41	1.93	0.28	1.29	0.75	3.50	0.90	4.20

The reproducibility study was conducted using two different reagent lots at three external sites. The protocol was run over 10 days, 2 runs per day, and 4 replicates per run for the sample pools, and 8 replicates per run for the negative and positive control materials. Reproducibility data was pooled across 3 sites.

Pool	# Repl.	Mean	Within Run		Between Run		Between Day		Total	
			SD	CV	SD	CV	SD	CV	SD	CV
Negative Control (plasma) (low negative)	480	0.11	0.004	NA	0.002	NA	0.01	NA	0.01	NA
Serum Pool 1 (low negative)	240	0.18	0.006	NA	0.002	NA	0.01	NA	0.01	NA
Serum Pool 2 (high negative)	240	0.75	0.01	1.3	0.01	1.4	0.01	1.3	0.02	2.3
Serum Pool 3 (low positive)	240	1.20	0.02	1.5	0.02	1.4	0.01	1.2	0.03	2.4
Serum Pool 4 (high positive)	240	6.67	0.10	1.5	0.11	1.6	0.08	1.2	0.17	2.5
Serum Pool 5 (high positive)	240	20.42	0.29	1.4	0.31	1.5	0.31	1.5	0.53	2.6
Positive Control (plasma) (moderate positive)	480	3.56	0.06	1.6	0.04	1.2	0.05	1.4	0.09	2.5

b. Linearity/assay reportable range:

Not Applicable

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

Reagent on-board stability studies were conducted. All data passed acceptance criteria. The on-board stability claim is for 60 days and calibration frequency for 21 days. Open-vial stability studies were also conducted for calibrators and controls. All lots tested met acceptance criteria. The open-vial claim for calibrators and for controls is 90 days at 2-8°C. Studies were also conducted to show that calibrators and controls can be kept on board the ADVIA Centaur system up to 24 hrs at room temperature.

Calibrators are prepared by spiking samples positive for *T. pallidum* into phosphate buffer containing BSA and preservatives to target concentrations of 0.65 and 8.0

The Negative Control is human negative plasma with a target index of <0.3. The positive control is prepared by spiking samples positive for *T. pallidum* into negative plasma to a target index of 4.0

d. Detection limit:

Samples with an Index Value < 0.90 are considered nonreactive for syphilis *T. pallidum a*ntibodies.

Samples with an Index Value ≥ 0.90 and < 1.10 are considered equivocal.

Samples with an Index Value ≥ 1.10 are considered reactive for syphilis *T. pallidum* antibodies.

e. Analytical specificity:

Cross reactivity

Special Populations

A total of 211 cord blood samples, samples from pregnant women (1st , 2nd, and 3rd trimester), samples from hospitalized patients, pediatric samples and transplant samples were tested using the ADVIA Centaur Syphilis assay and the predicate device (predicate device). Each sample was tested in singlicate using one lot of reagent on one system. Predicate device and Centaur Syphilis results were determined reactive, non-reactive or indeterminate/equivocal according to the respective result interpretation.

		Number of Reactive results	
Clinical Category	**Total Number Tested**	**Centaur Syphilis**	**Predicate device**
Cord Blood	18	1*	1
1st Trimester	24	1*	1
2nd Trimester	25	0	0
3rd Trimester	25	1*	1
Pediatric	48	0	0
Hospitalized	51	2*	2
Transplant Patients			
Heart	1	0	0
Kidney	4	0	0
Liver	6	0	0
Lung	9	0	0

* - positive on predicate device as well

A total of 265 specimens from 20 groups of potential cross-reactant disease states were assayed using the ADVIA Centaur Syphilis assay and the predicate device. These samples had a known activity to the potential cross reactant in each group of specimens which was determined by FDA-cleared methods and provided by the respective vendor. Each sample was tested in singlicate using

one lot of reagent. The predicate device and Centaur Syphilis results were determined reactive, non-reactive or indeterminate/equivocal according to the respective result interpretation method.

		Number of Reactive results	
Clinical Category	**Total Number Tested**	**Centaur Syphilis**	**Predicate device**
Lyme Disease	10	1*	1
Anti-Nuclear Antibody (ANA)	10	0	0
Rheumatoid Factor	10	0	0
HAMA	10	2*	2
Hepatitis A Infection (HAV) total	20	10*	10
Hepatitis A Infection (HAV) IgM	5	0	0
Hepatitis B Infection (HBV)	10	0	0
Hepatitis C Infection (HCV)	10	0	0
Human Immunodeficiency Virus (HIV)	11	0	0
Cytomegalovirus (CMV) IgG	10	0	0
Cytomegalovirus (CMV) IgM	5	0	0
Epstein-Barr Virus (EBV) IgG	10	0	0
Herpes Simplex Virus (HSV) IgG	10	5*	5
Rubella IgG	10	0	0
Rubella IgM	10	0	0
Toxoplasma IgG	10	1*	1
Toxoplasma IgM	10	0	0
Varicella Zoster Virus (VZV) IgG	10	2*	2
Lupus (SLE)	10	0	0
Drug users	20	3*	3
Myeloma patients	13	0	1
Flu Vaccine recipients	26	0	0
Hyper-IgG	5	0	0
Hyper-IgM	10	0	0

* - positive on predicate device as well.
* - all samples that demonstrated a positive result (with the exception of two HAV-positive samples) were also confirmed positive for syphilis by other tests (TPPA or RRP), indicating reactivity to *(T. Pallidum* antibodies) rather than cross reactivity.

Interference

Interference by endogenous substances was evaluated in the ADVIA Centaur syphilis assay. Serum pools were prepared at three levels (negative, low positive and high positive for *T. pallidum*). Each pool was spiked with an interferent to the levels indicated in the table below. There was no indication of interference up to the levels claimed. For positive samples all results demonstrated $\leq$ 10% change in index value, with the exception of gamma globulin at concentrations above 30 mg/dl.

Interferent	Tested concentration (up to)
Hemoglobin	500 mg/dL
Conjugated Bilirubin	40 mg/dL
Unconjugated Bilirubin	40 mg/dL
Intralipid	1000 mg/dL
Cholesterol, Total	400 mg/dL
Gamma Globulin	60 mg/dL (claim set at 30 mg/dL)
Protein, Total (HSA)	11 g/dL
Biotin	500 ng/mL

f. Assay cut-off:

The ADVIA Centaur Syphilis assay report results as reactive, non-reactive or equivocal based on Index Values. Index values are calculated based on a two point assay calibration curve. The cut-off value was established by evaluating negative and positive samples on the ADVIA Centaur Syphilis assay. The analysis showed a clear separation of negative and positive results using a cutoff index of 1. After establishment of the cut- off, an equivocal range of +/- 10% was set bracketing the cut-off value to encourage additional testing for index values that are close to the cut-off. The placement of the assay's cut-off and product performance were validated during clinical trials using receiver operating characteristic (ROC) analysis.

A receiver operating characteristic (ROC) analysis was performed to compare the diagnostic area under the curve of the ADVIA Centaur Syphilis assay to the IMMULITE 2000 assay. For validation of the cut off, the populations used for the analysis were the 806 apparently healthy subjects (negative diagnosis) and the 285 medically diagnosed subjects. The results show that the area under the curve is nearly identical for the two assays, and that the difference observed is not statistically significant ($p = 0.5282$). Results obtained using a cut-off of 1.0 in the ROC analyses demonstrated positive and negative percentage agreement is 97.9% and 99.4% respectively, meeting acceptance criteria.

2. Comparison studies:

a. Method comparison with predicate device:

The performance of the ADVIA Centaur Syphilis assay was evaluated at 3 clinical sites in geographically diverse locations. Percent agreement was determined by comparing the performance of the ADVIA Centaur SYPH assay to a commercially available syphilis assay. A total of 2108 samples were tested. These samples were from 474 apparently healthy subjects (including pediatrics), 285 medically-diagnosed syphilis samples, 124 samples reactive by previous laboratory testing (treponemal and non-

treponemal methods), 370 samples sent for routine syphilis testing, 339 samples from pregnant subjects (with or without reactive treponemal result), and 516 HIV-positive samples (with or without a reactive treponemal result). These samples were tested on the ADVIA Centaur system, split between 2 different test lots, and compared to a commercially available syphilis assay.

Percent Agreement: Total Study Population

The negative percent agreement of the ADVIA Centaur SYPH assay compared to the predicate device was 99.4% (1382/1391) with a 95% confidence interval (CI) of 98.8 to 99.7%.

The positive percent agreement of the ADVIA Centaur SYPH assay compared to the predicate device was 97.9% (700/715) with a 95% confidence interval (CI) of 96.6 to 98.8%

	Predicate Device			
ADVIA Centaur System	Reactive	Indeterminate	Nonreactive	Total
Reactive	700	1	6	707
Equivocal	1	0	3	4
Nonreactive	14	1	1382	1397
Total	715	2	1391	2108

Percent Agreement: Apparently Healthy Population

A population of 806 apparently healthy subjects was tested using the ADVIA Centaur SYPH assay and a commercially available syphilis assay. The performance of the ADVIA Centaur SYPH assay is shown in the following table:

Apparently Healthy Subjects	Reactive	Equivocal	Nonreactive	Total	Negative Percent Agreement
Pregnant	1 (0.3%)	0 (0.0%)	330 (99.7%)	332[a]	99.7% (329/330)
Pediatric	1 (1.3%)	0 (0.0%)	74 (98.7%)	75	98.6% (73/74)
Other[b]	3 (0.8%)	0 (0.0%)	396 (99.2%)	399	98.2% (389/396)
Total	5 (0.6%)	0 (0.0%)	801 (99.4%)	806	98.8% (791/801)

a One sample for the predicate device was indeterminate and was excluded.

b Other refers to samples from apparently healthy adults who are not pregnant.

Percent Agreement: Expected Positive Population

Samples from patient populations expected to test positive for syphilis were tested on the ADVIA Centaur and on a commercially available syphilis assay. These samples were from subjects found reactive by previous laboratory testing, and subjects who had been medically diagnosed with syphilis.

The positive percent agreement of the ADVIA Centaur SYPH assay to the comparator assay was 99.4% (535/538) with a 95% confidence interval (CI) of 98.4 to 99.9%. Percent negative agreement was 100% with a 95% CI of 85.2 to 100%

Expected Positive Subjects	Reactive	Equivocal	Nonreactive	Total	Positive Percent Agreement
TPPA-Reactive	271 (98.2%)	1 (0.4%)	4 (1.4%)	276	99.6% (271/272)
Medically Diagnosed	264 (92.6%)	0 (0.0%)	21 (7.4%)	285	99.2% (264/266)
Total	535 (95.4%)	1 (0.2%)	25 (4.5%)	561	99.4% (535/538)

Percent Agreement: Intended Use Population

Samples from patient populations expected to receive routine testing for syphilis (samples sent for routine testing and HIV-positive samples) were tested on the ADVIA Centaur and on a commercially available syphilis assay.

The negative percent agreement of the ADVIA Centaur SYPH assay compared to the comparator assay was 98.4% (568/577) with a 95% confidence interval (CI) of 97.1 to 99.3%.

The positive percent agreement of the ADVIA Centaur SYPH assay compared to the comparator assay was 98.2% (160/163) with a 95% confidence interval (CI) of 94.7 to 99.6%.

	Comparator Assay			
ADVIA Centaur System	Reactive	Indeterminate	Nonreactive	Total
Reactive	160	1	6	167
Equivocal	0	0	3	3
Nonreactive	3	0	568	571
Total	163	1	577	741

b. Matrix comparison:

A matrix study was performed to evaluate a variety of specimen tube types for compatibility with the ADVIA Centaur Syphilis assay. Serum glass was used as the control tube to which the following tubes were compared: Serum plastic (SST), K_2 EDTA plastic, lithium and sodium heparin plastic, sodium citrate plastic and ACD glass. Results showed that the negative samples stayed negative across all tube types. For positive samples, no bias was observed in all the other tube types except the ACD tube type. There was a slight increase in index value with negative samples in the ACD tube type so this tube type is not recommended for use with the assay.

3. Clinical studies:

a. Clinical Sensitivity:

Not Applicable (See Item 2a above)

b. Clinical specificity:

Not Applicable (See Item 2a above)

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

4. Clinical cut-off:

Not Applicable

5. Expected values/Reference range:

A total of 806 apparently healthy male and female subjects [including pregnant (332), pediatric (75), and adult/not pregnant (399)] were tested using the ADVIA Centaur Syphilis assay. Of these samples, 5 (0.6%) were reactive, 0 (0.0%) were equivocal, and 801 (99.4%) were nonreactive.

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.