

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION  
DECISION SUMMARY**

**A. 510(k) Number:**

K142826

**B. Purpose for Submission:**

To seek clearance for modification of the ADVIA Centaur Toxoplasma M (Toxo M) assay

**C. Measurand:**

*Toxoplasma* IgM antibodies

**D. Type of Test:**

Immunoglobulin Class-Capture Chemiluminescence Immunoassay

**E. Applicant:**

Siemens Healthcare Diagnostic, Inc.

**F. Proprietary and Established Names:**

ADVIA Centaur Toxoplasma M (Toxo M)

**G. Regulatory Information:**

1. Regulation section:

21CFR 866.3780; *Toxoplasma gondii* Serological Reagents

2. Classification:

Class II

3. Product code:

LGD; Enzyme Linked Immunosorbent Assay, *Toxoplasma gondii*

4. Panel:

83, Microbiology

## **H. Intended Use:**

### 1. Intended use(s):

The ADVIA Centaur Toxoplasma M (Toxo M) assay is an IgM antibody capture microparticle direct chemiluminometric in vitro diagnostic immunoassay intended for the qualitative detection of IgM antibodies to *Toxoplasma gondii* in serum or plasma (EDTA, heparin) using the ADVIA Centaur and ADVIA Centaur XP systems.

The ADVIA Centaur Toxo M assay is used to measure IgM antibody against *T. gondii* which is presumptive of an acute, recent, or reactivated toxoplasma infection. Any measurement of IgM antibody to *T. gondii* must be performed in conjunction with the determination of IgG antibody to *T. gondii*.

### 2. Indication(s) for use:

The ADVIA Centaur Toxoplasma M (Toxo M) assay is an IgM antibody capture microparticle direct chemiluminometric in vitro diagnostic immunoassay intended for the qualitative detection of IgM antibodies to *Toxoplasma gondii* in serum or plasma (EDTA, heparin) using the ADVIA Centaur and ADVIA Centaur XP systems.

The ADVIA Centaur Toxo M assay is used to measure IgM antibody against *T. gondii* which is presumptive of an acute, recent, or reactivated toxoplasma infection. Any measurement of IgM antibody to *T. gondii* must be performed in conjunction with the determination of IgG antibody to *T. gondii*.

### 3. Special conditions for use statement(s):

The detection of toxoplasma IgM in a given specimen, as determined by assays from different manufacturers, can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the toxoplasma IgM assay used. Values obtained with different assay methods cannot be used interchangeably. The reported IgM level cannot be correlated to an endpoint titer.

This assay is not intended for use in screening blood, plasma, or tissue donors. The effectiveness of this assay for use in screening blood, plasma, or tissue donors has not been established.

### 4. Special instrument requirements:

ADVIA Centaur and ADVIA Centaur XP

## **I. Device Description:**

The modified ADVIA Centaur Toxo M Assay is comprised of the following:

- ADVIA Centaur Toxo M Lite Reagent (10.0mL/pack)
  - Partially purified *T. gondii* antigen (~3 µg/mL) complexed with a mouse anti-*T. gondii* p30 monoclonal antibody (F(ab)2 fragment) labeled with acridinium ester in protein buffer with surfactant and preservatives
- ADVIA Centaur Toxo M Solid Phase (17.0mL/pack)
  - Mouse anti-human IgMµ monoclonal antibody (~24 µg/mL) covalently coupled to paramagnetic particles in protein buffer with surfactant and preservatives
- ADVIA Centaur Toxo M calibrators (600 µL/vial)
  - Defibrinated recalcified processed human plasma positive for toxoplasma IgM antibodies with preservatives.
- ADVIA Centaur Toxo M Controls (1.5mL/vial)
  - Defibrinated recalcified processed human plasma negative and positive for toxoplasma IgM antibodies with preservatives.

**J. Substantial Equivalence Information:**

1. Predicate device name(s):

ADVIA Centaur Toxoplasma M (Toxo M)

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

K010755

3. Comparison with predicate:

<b>Similarities</b>		
Item	Device: ADVIA Centaur Toxoplasma M K142826	Predicate: ADVIA Centaur Toxoplasma M K010755
<b>Intended Use</b>	The ADVIA Centaur Toxoplasma M (Toxo M) assay is an IgM antibody capture microparticle direct chemiluminometric <i>in vitro</i> diagnostic immunoassay intended for the qualitative detection of IgM antibodies to <i>Toxoplasma gondii</i> in serum or plasma (EDTA, heparin) using the ADVIA Centaur and ADVIA Centaur XP systems. The ADVIA Centaur Toxo M assay is used to measure IgM antibody against <i>T. gondii</i> which is presumptive of an acute, recent, or reactivated toxoplasma infection. Any measurement of	Same

<b>Similarities</b>		
Item	Device: ADVIA Centaur Toxoplasma M	Predicate: ADVIA Centaur Toxoplasma M
	K142826	K010755
	IgM antibody to <i>T. gondii</i> must be performed in conjunction with the determination of IgG antibody to <i>T. gondii</i> .	
<b>Sample Type</b>	Serum or plasma (EDTA, heparin)	Same
<b>Sample Volume</b>	10 µL	Same
<b>Instrument platforms</b>	ADVIA Centaur ADVIA Centaur XP	Same
<b>Calibration</b>	2-point calibration using Toxo M Calibrators	Same
<b>Capture Antibody (Solid Phase)</b>	Mouse anti-human IgMµ monoclonal antibody	Same
<b>Tracer (Lite Reagent)</b>	Toxoplasma p30 antigen bound to acridinium ester (via mouse anti- <i>T. gondii</i> p30 monoclonal antibody)	Same

<b>Differences</b>		
Item	Device	Predicate
	K142826	K010755
<b>Toxoplasma IgM Source (Calibrators, Controls)</b>	Cell culture supernatant of human anti-toxoplasma IgM monoclonal antibody-producing cells	Toxoplasma IgM positive human plasma pools
<b>Particle Resuspension</b>	Particle re-suspension with Wash 1 (phosphate buffered saline)	Particle re-suspension with water
<b>Lite Reagent Conjugate</b>	Ab format = F(ab) <sub>2</sub> fragment Ab Concentration = 12.5 ng/ mL Conjugate Loading Ratio = 18:1	Ab format = Whole IgG Ab Concentration = 30 ng/mL Conjugate Loading Ratio = 30:1
<b>Solid Phase Buffer</b>	Buffer: Tricine (pH = 8.0) NaCl: 300 mM Surfactant: Tween-20 = 2.2g/L Blocker: sm-BSA = 10.0 g/L Mouse IgG: 100 mg/L EDTA: 0.7 g/L	Buffer: Tris (pH =8.0) NaCl: 150 mM Surfactant: CHAPS = 0.1 g/L Blocker: Gelatin = 22.2 g/L Mouse IgG: 25 mg/L EDTA: none
<b>Claimed Measuring Range</b>	0.10–10.00 Index	0.10–40.00 Index

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

The following recognized standards from Clinical Laboratory Standards Institute (CLSI) were used as a basis of the study procedures described in this submission:

- Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline– Second Edition (CLSI EP05-A2, 2004; Recognition No. 7-110)
- Interference Testing in Clinical Chemistry; Approved Guideline – Second Edition (CLSI EP07-A2, 2005; Recognition No. 7-127)
- Stability testing of in vitro diagnostic reagents (European Committee for Standardization EN 13640:2002; Recognition No. 7-84)
- Medical devices – Application of risk management to medical devices (ANSI/AAMI/ISO 14971:2007/(R)2010; Recognition No. 5-70)

#### **L. Test Principle:**

The ADVIA Centaur Toxo M assay is an immunoglobulin class-capture sandwich immunoassay using direct, chemiluminometric technology. The anti-human IgM monoclonal antibody is covalently coupled to paramagnetic particles in the Solid Phase. In the Lite Reagent, the *T. gondii* antigen is complexed with an anti-p30 monoclonal antibody (F(ab)<sub>2</sub> fragment) labeled with acridinium ester. Antibody-antigen complexes will form if toxoplasma IgM is present in the sample.

ADVIA Centaur systems automatically perform the following steps for the Toxo M assay:

- Dispenses 10 µL of sample into a cuvette.
- Dispenses 340 µL of Solid Phase and incubates the mixture for 18 minutes at 37°C.
- Separates the Solid Phase from the mixture and aspirates the unbound reagent.
- Washes the cuvette with Wash 1.
- Dispenses 200 µL Lite Reagent and incubates the mixture for 18 minutes at 37°C.
- Separates the Solid Phase from the mixture and aspirates the unbound reagent.
- Washes the cuvette with Wash 1.
- Dispenses 300 µL each of Acid Reagent and Base Reagent to initiate the chemiluminescent reaction.
- Reports results according to the selected option, as described in the system operating instructions or in the online help system

A direct relationship exists between the amount of toxoplasma IgM activity present in the patient sample and the amount of relative light units (RLUs) detected by the system. A result of reactive (positive) or nonreactive (negative) is automatically determined using an Index Value.

#### **M. Performance Characteristics (if/when applicable):**

##### 1. Analytical performance:

###### *a. Precision/Reproducibility:*

Precision of the modified ADVIA Centaur Toxo M assay was evaluated according to CLSI EP05-A2. The study was conducted for 20 days with 2 runs per day using 1 lot of reagents and 2 instruments. The materials tested consisted of lot-specific

calibrators, one lot of controls, and four patient samples (low positive, moderate positive, high positive). For each run, samples were tested in duplicate. Within-run CV and total CV were calculated. The predetermined acceptance criteria of  $\leq 10\%$  (with in run CV) and  $\leq 15\%$  (total CV) for specimens ranging from 0.4 to 0.9 Index and  $\leq 8\%$  (with in run CV) and  $\leq 12\%$  (total CV) for specimens  $\geq 1.0$  index. The within-run and total precision obtained with the modified ADVIA Centaur Toxo M reagents on the ADVIA Centaur (XP) system meets acceptance criteria.

*b. Linearity/assay reportable range:*

The modified ADVIA Centaur Toxo M assay is a qualitative assay. Linearity studies are not applicable. The assay reportable range is 0.10–10.00 Index.

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

**Stability:**

Stability studies were performed to establish the in-use stability claims (onboard stability and calibration interval) for modified ADVIA Centaur Toxo M reagents kept onboard the ADVIA Centaur (XP) system. The onboard stability of the ADVIA Centaur Toxo M reagents is 28 days with a calibration interval of 14 days. The reagents and calibrators are stable until the date printed on the box label when stored at 2-8°C. The claimed shelf-life of the modified ADVIA Centaur Toxo M reagents and calibrators are 14 months.

The predetermined acceptance criteria were met and the results claim the 28-day onboard stability claim with the 14 day calibration interval for the for modified ADVIA Centaur Toxo M reagents on the ADVIA Centaur (XP) system.

*d. Detection limit:*

Not applicable

*e. Analytical specificity:*

**Cross-Reactivity:**

The performance of the modified ADVIA Centaur Toxo M assay was evaluated in the presence of 204 potentially cross-reacting substances using two lots of reagent. The results are summarized in Table 1 below.

Table 1. Summary of Results from Cross-Reactivity Study

Sample Type	Lot 1				Lot 2		
	N	Neg	Equivocal	Pos	Neg	Equivocal	Pos
Anti-mitochondrial antibody (AMA)	15	15	0	0	15	0	0
Anti-nuclear antibody (ANA)	19	19	0	0	19	0	0
Human anti-mouse antibody (HAMA)	25	24	1	0	24	1	0
Rheumatoid factor (RF)	29	29	0	0	29	0	0
Multiple Myeloma IgM	13	13	0	0	13	0	0
anti-Cytomegalovirus (CMV) IgM	14	14	0	0	14	0	0
anti-Epstein Barr (EBV) IgM	16	16	0	0	16	0	0
anti-Herpes Simplex (HSV) IgM	20	20	0	0	20	0	0
anti-Measles (Rubeola) IgM	10	10	0	0	10	0	0
anti-Parvovirus B19 IgM	10	10	0	0	10	0	0
anti-Syphilis IgM	12	12	0	0	12	0	0
anti-Varicella Zoster (VZV) IgM	21	21	0	0	21	0	0
<b>Total</b>	<b>204</b>	<b>203</b>	<b>1*</b>	<b>0</b>	<b>203</b>	<b>1*</b>	<b>0</b>

\*This sample was equivocal in lot 1 and lot 2 when tested with the modified ADVIA Centaur Toxo M assay and negative when tested with the unmodified predicate ADVIA Centaur Toxo M assay.

The predetermined acceptance criteria were no false positive results with any interfering disease states tested. The results demonstrated no false positives results found, the results of the cross reactivity study were acceptable.

**Endogenous Interference:**

The performance of the modified ADVIA Centaur Toxo M assay was evaluated according to CLSI EP07-A2 in the presence of low and high levels of endogenous substances (hemoglobin, conjugated and unconjugated bilirubin, triglycerides (intralipids), and total protein). Samples in 3 matrices (serum, LiHep plasma, and EDTA plasma) from 3 donors were spiked with toxoplasma IgM at three levels (negative, low positive and high positive), then the potentially-interfering endogenous substance were added. The results are pooled and summarized in Table 2 below.

The bias was calculated as follows for the low positive and high positive samples:

$$\%Bias = \frac{(Index\ of\ Interferent\ -Spiked\ Sample - Index\ of\ Diluent\ -Spiked\ Sample)}{Index\ of\ Diluent\ -Spiked\ Sample}$$

Table 2. Toxo IgM Results in the Presence of Endogenous Substance at the Indicated Levels

Endogenous Substance	Endogenous Substance Levels	Bias		
		Negative Toxo IgM	Low Positive Toxo IgM	High Positive Toxo IgM
Hemoglobin	250 mg/dL, 500 mg/dL	Negative	<10%	<10%
Triglycerides (Intralipid)	500 mg/dL, 1000 mg/dL	Negative	<10%	<10%
Unconjugated Bilirubin	20 mg/dL, 40 mg/dL	Negative	<10%	<10%
Conjugated Bilirubin	30 mg/dL, 60 mg/dL	Negative	<10%	<10%
Protein	3 g/dL, 12 g/dL	Negative	<10%	<10%

For nonreactive specimens, all samples (serum, LiHep plasma, and EDTA plasma) remained negative in the presence of interferents at the levels tested. For both low positive and high positive toxoplasma IgM spiking levels in each matrix (serum, LiHep plasma, and EDTA plasma) and across matrices, the calculated bias was below 10% for all interferents: therefore the predefined acceptance criteria of nonreactive samples must remain nonreactive in the presence of interferents and for reactive samples, the grand mean % difference in Index (across all 3 matrices) must be less than 10% were met.

*f. Assay cut-off:*

The modifications to the ADVIA Centaur Toxo M assay did not alter the cutoff (as reported in K010755).

- Samples with a calculated value of less than 0.9 Index are considered nonreactive (negative).
- Samples with a calculated value between 0.9 and 0.99 Index are considered equivocal.
- Samples with a calculated value greater than or equal to 1.0 Index are considered reactive (positive).

2. Comparison studies:

*a. Method comparison with predicate device:*

The method comparison study was conducted by assaying 1111 clinical specimens. The testing included: 1) Panel specimens (n = 126) containing well characterized clinical specimens and 2) Patient specimens (n = 985) consisting of prenatal specimens, prospective, and characterized specimens with known clinical status from various locations, including the US and France.

Specimens were assayed in single replicate on ADVIA Centaur (XP) with the modified ADVIA Centaur Toxo M assay and the unmodified predicate ADVIA Centaur Toxo M assay.

Table 3 shows results obtained for the 985 patient specimens. The composition of the patient specimens is as follows:

- 98 prenatal specimens (Source: US)
- 164 prenatal specimens (Source: France)
- 507 prospective (Source: US)
- 187 characterized (Source: US)
- 25 characterized (Source: France)
- 4 characterized (Source: in-house)

Table 3. Percent Agreement (Modified vs Unmodified Toxo M): Patient specimens

		Unmodified Predicate ADVIA Centaur Toxo M			Total
		Reactive	Equivocal	Nonreactive	
Modified ADVIA Centaur Toxo M	Reactive	163	0	2	165
	Equivocal	1	0	1	2
	Nonreactive	1	0	817	818
	Total	165	0	820	985

The two specimens that produced equivocal results in the modified ADVIA Centaur Toxo M assay were removed from the analysis. Therefore the total numbers of reactive and non-reactive specimens were 164 and 819, respectively. The positive percent agreement was 99.4% (163/164) with a 95% confidence interval (CI) of 96.65%–99.98%, and the negative percent agreement was 99.8% (817/819) with 95% CI of 99.12%–99.97%.

Table 4 shows the results obtained for panel specimens. The composition of the panel specimens is as follows:

- 100 members of the CDC Toxoplasma performance panel (includes 35 positives)
- 22 members of a Toxoplasma performance panel (Source: Germany)
- 4 members of the Toxo M QC panel (Source: in-house)

Table 4. Percent Agreement (Modified vs Unmodified Toxo M): Panel Specimens

		Unmodified Predicate ADVIA Centaur Toxo M			Total
		Reactive	Equivocal	Nonreactive	
Modified ADVIA Centaur Toxo M	Reactive	53	1	0	54
	Equivocal	1	0	0	1
	Nonreactive	1	0	70	71
	Total	55	1	70	126

The two specimens that produced equivocal results (one specimen each by the modified and unmodified assay) were removed from the analysis. Therefore the total number of reactive specimens was 54. The number of non-reactive specimens was not affected. The positive percent agreement was 98.1% (53/54) with 95% CI of 90.11%–99.95%, and the negative percent agreement was 100.0% (70/70) with 95% CI of 94.87%–100.00%.

The acceptance criteria as defined by the sponsor, for the modified ADVIA Centaur Toxo M assay compared to the unmodified predicate ADVIA Centaur Toxo M assay was as follows:

- Positive Percent Agreement  $\geq 97.21\%$
- Negative Percent Agreement  $\geq 98.56\%$

These results met the acceptance criteria for both patient specimens and panel specimens.

*b. Matrix comparison:*

Not applicable

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:

The unmodified predicate ADVIA Centaur Toxo M assay was designed to have a positive cutoff of 1.00 Index. The cutoff for the unmodified predicate ADVIA Centaur Toxo M assay was originally established by running a known population of 1295 samples using the unmodified predicate ADVIA Centaur Toxo M assay and adjusting the cutoff Index value between known positives and non-positives to 1.0.

The modifications to the current modified ADVIA Centaur Toxo M assay did not alter the cutoff.

Expected values/Reference range:

The incidence of toxoplasmosis varies considerably by the geographic location and age of patient as indicated in Table 5.

Table 5. Toxoplasmosis Incidence As Reported in Literature

<b>Location</b>	<b>Seroprevalence Rate</b>
France, Italy	50–85%, by region
Germany	20–72%, by region
United Kingdom	20%
Japan	24%
Africa	20–65%, by country
S. America	36–82%, by country
N. America	8–38%, by region

**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.