

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

A. 510(k) Number: K080869

B. Purpose for Submission: Initial Premarket Notification

C. Measurand: Human IgG class antibodies to *Toxoplasma gondii* (Toxo IgG) in serum

D. Type of Test: Chemiluminescent immunoassay for qualitative and quantitative determination of *Toxo IgG* in diagnosis of *Toxoplasma gondii* infection

E. Applicant: Beckman Coulter, Inc.

F. Proprietary and Established Names: Access Toxo IgG Assay, Access Toxo IgG Calibrators, Access Toxo IgG QC

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
LGD	Class II	866.3780	Microbiology (83)

H. Intended Use:

The Access Toxo IgG assay is a paramagnetic-particle, chemiluminescent immunoassay for the qualitative and quantitative determination of IgG antibodies to *Toxoplasma gondii* in human serum using the Access Immunoassay Systems. The Access Toxo IgG assay aids in the diagnosis of *Toxoplasma gondii* and may be used to assess the immune status of pregnant women.

This product is not FDA cleared/approved for the screening of blood of plasma donors. Assay performance characteristics have not been established for immunocompromised or immunosuppressed patients, cord blood, neonatal specimens of infants.

3) Special conditions for use statement(s): For Prescription use only

4) Special instrument requirements: see I

I. Device Description: The device consists of three different components: Access Toxo IgG Assay reagents, Toxo IgG Calibrators, and Toxo IgG QC. The assay is performed for the quantitative and qualitative determination of anti-*Toxoplasma gondii* IgG in human serum and is read using the Access Immunoassay Analyzers (Access,

Access 2, Synchron LXi 725, UniCel DxC 600i, UniCel DxI 600 and UniCel DxI 800).

J. Substantial Equivalence Information:

a) Predicate device name (s):

Beckman Coulter, Inc.- Access Toxo IgG Assay, Calibrators, QC (Part Numbers: 34450, 34455, 34459)
 Bio-Rad Laboratories, Inc. – Access Toxo IgG Assay on the Access Immuno Assay
 Abbot Laboratories - AxSYM Toxo IgG Antibody Assay (Comparator)

b) Predicate K numbers (s):

Beckman Coulter, Inc. - K032162

Bio-Rad Laboratories, Inc. - K951495

Abbot Laboratories - K954575 (Comparator)

Comparison with predicate: Three devices were listed as predicate devices. The AxSYM Toxo IgG Antibody Assay from Abbot Laboratories was used as a reference method for clinical data evaluation and calculation of positive and negative percent agreement. The following Table shows the similarities and differences between Beckman and Abbot Toxo IgG Assay

Similarities	Predicate Device Abbot Laboratories	Beckman Coulter, Inc.
Attribute	AxSYM Toxo IgG - K954575	New Access Toxo IgG (P/N A31588)
Assay Methodology	Microparticle solid-phase, two-step enzyme immunoassay.	Microparticle solid-phase, two-step enzyme immunoassay.
Intended Use	Qualitative and quantitative measurement of IgG antibodies to <i>Toxoplasma gondii</i> .	Qualitative and quantitative determination of IgG antibodies to <i>Toxoplasma gondii</i> .
Solid Phase	Microparticles coated with partially-purified <i>T. gondii</i> antigen (RH strain from HeLa culture).	Microparticles coated with partially-purified <i>T. gondii</i> antigen (RH strain).
Conjugate	Alkaline phosphatase-labeled anti-human IgG antibody.	Alkaline phosphatase-labeled anti-human IgG antibody.
Calibrators	Six liquid re-calcified plasma	Six liquid defibrinated plasma

	calibrators (one non-reactive, five reactive).	calibrators (one non-reactive, five reactive).
Controls	Two liquid re-calcified plasma controls (one non-reactive, one reactive).	Two liquid defibrinated plasma controls (one non-reactive, one reactive).
Differences		
Abbot Laboratories		Beckman Coulter Inc
Attribute	AxSYM Toxo IgG - K954575	Beckman New Access Toxo IgG (P/N A31588)
Reporting Methodology	Enzyme immunoassay employing fluorescent detection.	Enzyme immunoassay employing chemiluminescent detection.
Sample Type	Human serum or plasma (EDTA, heparin, sodium citrate).	Human serum.
Solid Phase Capture Mechanism	Microparticles captured onto glass fibers for washing and signal detection.	Paramagnetic microparticles retained in a magnetic field for washing and signal detection.
Conjugate Antibody	Goat polyclonal anti-human IgG antibody.	Mouse monoclonal anti-human IgG antibody.
Substrate	4-methylumbelliferyl phosphate	Lumi-Phos 530 (dioxetane-based substrate)
System Wash Solution(s)	Matrix Cell Wash - TRIA-buffered saline with sodium azide and antimicrobials. Line Diluent - phosphate buffer with sodium azide and antimicrobials.	Access Wash Buffer II - TRIS-buffered saline, Tween 20 surfactant, sodium azide and ProClin 300.
Calibration method	Six-point calibration curve or two-point master curve.	Six-point calibration curve.
Calibrator Reference	WHO 2 nd International Standard Preparation for Anti- <i>Toxoplasma</i> Serum.	WHO 3rd International Standard Preparation for Anti- <i>Toxoplasma</i> Serum.
Calibration Validity	Indefinite (required with new reagent lot or upon quality control failure).	28 Days
Reactive Cut-off	>or = 3 IU/mL	>or = 10.5 IU/mL
Gray Zone	2 - 3 IU/mL	7.5 - 10.5 IU/mL

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

L. Test Principle: The Access Toxo IgG assay is an enzyme immunoassay using an indirect technique. A sample is added to a reaction vessel with paramagnetic particles coated with *Toxoplasma gondii* membrane antigen. Specific antibodies present in the sample bind to the antigen. After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away. Alkaline phosphatase-conjugated monoclonal anti-human IgG antibody is then added and attaches to the IgG antibodies captured on the particles. A second separation and wash step removes unbound conjugate. Then, the chemiluminescent substrate Lumi-Phos* 530 is added to the vessel and light generated by the reaction is measured with a luminometer. The light production is directly proportional to the concentration of Toxo IgG antibody in the sample. The amount of analyte in the sample is determined from a stored, multi-point calibration curve.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:* Reproducibility/repeatability of the Access Toxo IgG Assay was estimated at one internal (Site 3) and two external sites (Sites 1 and 2). Each site performed one run per day over seven test days. Nine serum samples were run in replicates of five in each run. Combined results for each serum sample from all three sites are presented in the table below. The observed average total %CV was 12.6% with daily calibration and 15.3% using a stored calibration.

Combined Results											
Sample	N	Mean Dose (IU/mL)	Daily Calibration				Stored Calibration				
			Intra-assay %CV	Inter-assay %CV	Inter-site %CV*	Total %CV *	Mean Dose (IU/mL)	Intra-assay %CV	Inter-assay %CV	Inter-site %CV *	Total %CV *
A001	105	3.70	13.5	6.9	13.0	20.0	3.73	12.1	5.7	6.9	15.1
A002	105	8.88	7.7	2.5	9.4	12.5	8.73	6.9	2.7	15.6	17.3
A010	105	12.7	8.1	2.4	10.5	13.5	12.4	6.3	0.3	15.2	16.4
A003	105	13.6	8.1	3.1	9.8	13.1	13.4	6.6	0.7	15.6	17.0
A004	105	18.7	7.6	3.1	6.3	10.3	18.3	7.8	1.8	11.8	14.3
A009	105	43.8	6.1	1.6	8.6	10.7	43.0	5.4	1.1	12.6	13.7
A005	105	89.4	7.2	3.4	5.1	9.5	88.9	7.1	3.9	12.3	14.8
A006	105	246.4	7.2	3.2	7.7	11.0	250.0	7.9	3.5	10.6	13.7
A007	105	286.7	7.0	1.3	10.6	12.7	292.5	8.8	2.9	12.8	15.8

*The inter-site and total estimates also include contributions from inter-lot variation.

b. Linearity/assay reportable range:

Dilution Recovery (Linearity)

Recovery with WHO Standard: To calibrate the Access Toxo IgG assay to the 3rd

International Standard for Anti-Toxoplasma serum (TOXM 1-85), five dilutions of the Third International Standard for Anti-Toxoplasma Serum (TOXM) were tested in duplicate in a single run. The recovery performance is presented in the following Table. The average recovery for all dilutions is 93.5%.

WHO Standard Recovery

Expected Dose (IU/mL)	Observed Dose (IU/mL)	Mean Observed Dose (IU/mL)	Recovery
6.0	5.8 6.0	5.9	98.3%
30.0	26.6 25.2	25.9	86.3%
60.0	65.4 64.8	65.1	108.5%
120.0	117.0 114.4	115.7	96.4%
240.0	183.6 191.1	187.4	78.1%

Linearity with High Patient Samples: Four highly reactive sera were diluted from 1/1.5 to 1/32 in Access Toxo IgG Calibrator, S0. These dilutions were tested in quadruplicate and the results were compared to expected values based on the neat sample determination (eight replicates). The mean recovery for the four sera was 100% and ranged from 93.8% to 109.1%.

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

Access Toxo Calibrators(A31589): The Access Toxo calibrators are traceable to the WHO 3rd International Standard Preparation for Anti-*Toxoplasma* Serum (TOXM) based on EN ISO 17511. The calibrators are provided at six levels- zero(S0), and approximately 10.5 (S1), 50 (S2), 120(S3), 240(S4), and 450(S5) IU/mL. The recommended open use life of the Calibrators is 90 days.

Access Toxo Quality Controls(A31590): There are two controls: 1) QC1- Pooled human defibrinated plasma negative (non-reactive) for anti-*T.gondii* IgG; 2) QC2- Spiked human defibrinated plasma containing high levels of anti-*T.gondii* IgG positive (reactive). The target range for QC1 is <2 IU/mL; the target range for QC2 is 20-30 IU/mL. The recommended open use life of the Controls is 90 days.

d. *Analytical sensitivity:*

(i) *Limit of Blank (LoB):* The LoB was determined with three serum samples in 60 replicates (level of anti-Toxoplasma IgG approaching 0 IU/mL in each serum sample) and one lot of the S0 calibrator with two lots of Access Toxo IgG reagents (P1 and P2). The testing was conducted with one Access and one Access 2 Analyzer. The

LoB for the Access Toxo IgG is estimated at 0.10 IU/mL.

(ii) *Limit of Detection (LoD)*: Four low level (non-reactive) serum samples were tested in 60 replicates with two reagent lots. The testing was conducted with one Access and one Access 2 Analyzer. The LoD for the Access Toxo IgG is estimated at 0.40 IU/mL.

(iii) *Limit of Quantitation (LoQ)*: To estimate the LoQ for the Access Toxo IgG, two samples with established levels of anti-Toxoplasma IgG reactivity was tested over 5 days in one run of 5 replicates per day. The testing was conducted with two reagent lots on a single Access Analyzer. The mean LoQ for the Access Toxo IgG is 3.2 IU/mL.

e. *Analytical specificity/Interference*: A study was conducted to investigate potential cross-reactivity with immunoglobulins resulting from exposure to other infectious agents that can produce symptoms similar to *Toxoplasma* infection (CMV, Epstein-Barr virus, HIV, HSV-1, HSV-2, malaria, measles, rubella VZV, mumps and *Treponema*). In addition, interference due to heterophilic antibodies (HAMA), abnormal immune system conditions (myeloma, rheumatoid factor, ANA) and influenza vaccine was evaluated. A total of 311 samples were tested. Nine samples (2.9%, including HIV, HSV-2 IgG, HAMA, mumps, syphilis) that were non-reactive by another commercially available assay were equivocal or reactive in the Access Toxo IgG Assay. The observed performance with each condition is presented in the following Table:

Condition	Number tested	Equivocal or Reactive in Access Toxo IgG
ANA	14	0
CMV IgG	8	0
EBV IgG	13	0
HAV Ab	10	0
HBV (HBsAg)	11	0
HCV Ab	14	0
HIV	43	2
HSV-1 IgG	3	0
HSV-2 IgG	8	2
Malaria	7	0
Measles IgG	13	0
Myeloma IgG	12	0
Rheumatoid Factor	15	0
Rubella IgG	12	0
VZV IgG	11	0
HAMA/Heterophilic Antibody	20	2
Mumps IgG	65	2
Influenza (vaccine recipients)	23	0
Syphilis	43	1

The Access Toxo IgG Assay was not significantly affected by the presence of 300 mg/L bilirubin (100 mg/L free + 200 mg/L conjugated), 30 g/L triolein (triglycerides), 90 g/L albumin or 20 g/L hemoglobin.

f. Assay cut-off: The assay cutoff was established by an evaluation of 959 non-reactive and 1091 reactive samples characterized by another enzyme immunoassay method. Cutoff for the Access Toxo IgG assay was determined to be 10.5 IU/mL based on receiver operating characteristic (ROC) curves.

- All test samples < 7.5 IU/mL are considered non-reactive for the presence of *Toxoplasma gondii* IgG antibodies.
- Sample results ≥ 7.5 IU/mL and < 10.5 IU/mL are considered equivocal for the presence of *Toxoplasma gondii* IgG antibodies.
- All test samples ≥ 10.5 IU/mL are considered reactive for the presence of *T. gondii* IgG antibodies.

2. Comparison studies:

a. Method comparison with reference method:

Abbot AxSYM Toxo IgG Antibody Assay

b. Matrix comparison: NA

3. Clinical studies:

a. Clinical Sensitivity: see below

b. Clinical specificity: see below

Performance Characteristics

Summary of Clinical Studies

Method Comparison: Studies comparing the performance of the Access Toxo IgG assay with the Abbott AxSYM Toxo IgG method were conducted at one external site in south-central France (Site 1), one external site in the northeastern United States (Site 2) and at the manufacturer's site (Site 3). The external sites tested samples collected from their own routine prenatal screening population as well as specimens from males and non-pregnant females that had Toxo IgG testing ordered. Two clinical sample suppliers provided the routine Toxo IgG test specimens for the U.S. site. The manufacturer tested prenatal specimens collected at three hospitals in north-central France.

Method agreement results for the prospective (fresh) and retrospective (frozen) collections by test site are presented in the following two Tables.

Retrospective/Prospective Patient Population

Comparison EIA			+	+	+	EQV	EQV	EQV	-	-	-
n	Access		+	EQV	-	+	EQV	-	+	EQV	-
Site 1	406	Frozen	154	3	2	3	3	4	0	2	235
Site 2	28	Fresh	4	0	1	0	0	0	0	0	23
Site 2	433	Frozen	190	2	0	4	6	0	1	0	230
Site 3	558	Frozen	356	2	0	2	0	0	1	1	196

Agreement Table for Retrospective/Prospective Patient Population

			Positive Agreement (%)	95% Conf. Int.	Negative- Agreement (%)	95% Conf. Int.
Site 1	406	Frozen	94.5	89.8 - 97.4	97.9	95.2 – 99.3
Site 2	28	Fresh	80.0	28.5 – 99.5	100	85.3 - 100
Site 2	433	Frozen	99.0	96.3 – 99.9	97.9	95.1 – 99.3
Site 3	558	Frozen	99.4	98.0 – 99.9	98.0	94.9 – 99.4

Method agreement results for the prenatal and diagnostic populations (male and female) by test site are presented in the following two Tables.

Pregnant/Non-Pregnant Population

Comparison EIA			+	+	+	EQV	EQV	EQV	-	-	-
n	Access		+	EQV	-	+	EQV	-	+	EQV	-
Site 1	229	Pregnant	37	3	0	2	2	2	0	1	182
Site 2	173	Pregnant	13	0	1	0	0	0	1	0	158
Site 3	558	Pregnant	356	2	0	2	0	0	1	1	196
Site 1	76	Female	47	0	2	1	0	2	0	0	24
Site 1	101	Male	70	0	0	0	1	0	0	1	29
Site 2	180	Female	97	2	0	4	6	0	0	0	71
Site 2	108	Male	84	0	0	0	0	0	0	0	24

Agreement Table for Pregnant/Non-Pregnant Population

			Positive Agreement (%)	95% Conf. Int.	Negative Agreement (%)	95% Conf. Int.
Site 1	229	Pregnant	88.1	74.4 – 96.0	98.4	95.3 – 99.7
Site 2	173	Pregnant	92.9	66.3 – 99.8	99.4	96.5 – 100
Site 3	558	Pregnant	99.4	98.0 – 99.9	98.0	94.9 – 99.4
Site 1	76	Female	92.2	81.1 – 97.8	96.0	79.7 – 99.9
Site 1	101	Male	100	94.9 – 100	96.7	82.8 – 99.9
Site 2	180	Female	98.0	92.9 – 99.8	94.7	86.9 – 98.5
Site 2	108	Male	100	95.7 - 100	100	85.8 - 100

CDC *Toxoplasma* 1998 Human Serum Panel

The Access Toxo IgG Assay exhibited 100% positive and negative agreement with the 100-member CDC *Toxoplasma* 1998 Human Serum Panel.

4. Clinical cut-off: As described in Assay cut-off.
5. Expected values/Reference range: The worldwide prevalence of *Toxoplasma* infection varies widely, influenced by such factors as environmental conditions, sanitation, dietary practices and companion and farm animal contact. Local prevalence is also influenced by population age and immigration patterns. Published estimates of seroprevalence for selected population in Europe, Asia, Africa and south America range from <5 -90% while a recent study estimate the overall United States prevalence to be approximately 22%.

Due to this substantial variability, it was recommended to establish prevalence expectations based on its own specific patient population.

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. The Access Toxo IgG Assay, Calibrators and QC (Part numbers: A31588, A31589, A31590) on the Access Immunoassay Systems are substantially equivalent to the AxSYM Toxo IgG Assay and the Access Toxo IgG Assay, Calibrators and QC (Part numbers: 34450, 34455, 34459) for the quantitative and qualitative determination of IgG antibodies to *Toxoplasma gondii* in human serum.