



### AccuDiag™ Toxoplasma gondii IgG (Toxo IgG) ELISA Kit

REF 1101

PIC PB1101ZY1

IVD See External Label 8°C 96 Tests

Toxoplasma gondii IgG (Toxo IgG) ELISA	
Principle	Indirect ELISA
Detection	Qualitative
Sample	10 µL serum
Incubation Time	60 minutes
Sensitivity	95.3%
Specificity	100%
Shelf Life	12 Months from the manufacturing date

#### PRODUCT FEATURES

- Very easy to use with little training
- Highly specific and consistent Assay
- Provides accurate results quickly
- Reading of results both visually and as absorbance data

#### INTENDED USE

The Diagnostic Automation/Cortez Diagnostics, Inc. Toxoplasma (Toxo) IgG Enzyme-Linked Immunosorbent Assay (ELISA) is intended for the detection and quantitative determination of IgG antibody to Toxoplasma in human sera. This product is not FDA cleared (approved) for use in testing (ie, screening) blood or plasma donors. **For in vitro diagnostic Use. High complexity test.**

#### SIGNIFICANCE AND SUMMARY

Toxoplasma is a coccidian parasite initially isolated in 1908 from a North African rodent. Since then, the organism has been found in many species of birds, reptiles and mammals.<sup>1</sup>

Man is infected with Toxoplasma from various suspected sources: ingestion of infected meat, especially mutton and pork, or ingestion of soil contaminated by oocysts from domestic and feral cat.<sup>2</sup> Transmission by organ transplant, transfusion or activation of quiescent infections is also documented. Congenital Toxoplasmosis is a disease with an extraordinarily wide range of manifestations; so wide in fact, that it must be considered in the differential diagnosis of nearly all types of obscure illness occurring during infancy.<sup>3</sup>

Because symptoms are sometimes nonspecific (i.e., anemia, splenomegaly, jaundice, fever, hepatomegaly, adenopathy and vomiting), congenital Toxoplasmosis is easily misdiagnosed on the clinical grounds, even in sick infants who have the generalized form of the disease.<sup>4</sup>

Toxoplasmosis must also be considered in the differential diagnosis in any immunosuppressed patient who has clinical or laboratory evidence of damage to the central nervous system.<sup>5</sup>

The organism is one of the most common latent infectious agents of man throughout the world.<sup>6</sup>

The Diagnostic Automation, Inc. Toxoplasma IgG ELISA kit provides all the necessary reagents for the rapid quantitation of Toxoplasma IgG antibody in human sera.

The sensitivity, specificity, and reproducibility of enzyme-linked immunosorbent assays is comparable to other serological tests for antibody, such as immunofluorescence, complement fixation, hemagglutination and radioimmunoassay.<sup>11, 12, 13</sup>

#### ASSAY PRINCIPLE

Enzyme-Linked Immunosorbent Assays (ELISA) rely on the ability of biological materials (i.e., antigens) to adsorb to plastic surfaces such as polystyrene (solid phase). When antigens bound to the solid phase are brought into contact with a patient's serum, antigen specific antibody, if present, will bind to the antigen on the solid phase forming antigen-antibody complexes. Excess antibody is removed by washing. This is followed by the addition of goat anti-human IgG conjugated with horseradish peroxidase which then binds to the antibody-antigen complexes. The excess conjugate is removed by washing, followed by the addition of Chromogen/Substrate, tetramethylbenzidine (TMB). If specific antibody to the antigen is present in the patient's serum, a blue color develops. When the enzymatic reaction is stopped with 1N H<sub>2</sub>SO<sub>4</sub>, the contents of the wells turn yellow. The color, which is indicative of the concentration of antibody in the serum, can be read on a suitable spectrophotometer or ELISA microwell plate reader.<sup>7, 8, 9, 10</sup>

#### SPECIMEN COLLECTION AND PREPARATION

1. Handle all blood and serum as if capable of transmitting infectious agents.
2. Optimal performance of the Diagnostic Automation, Inc. ELISA kit depends upon the use of fresh serum samples (clear, non-hemolyzed, nonlipemic, non-icteric). A minimum volume of 50 µL is recommended, in case repeat testing is required. Specimens should be collected aseptically by venipuncture.<sup>18</sup> Early separation from the clot prevents hemolysis of serum.
3. Store serum between 2 and 8°C if testing will take place within two days. If specimens are to be kept for longer periods, store at -20°C or colder.



Do not use a frost-free freezer because it may allow the specimens to go through freeze-thaw cycles and degrade antibody. Samples that are improperly stored or are subjected to multiple freeze-thaw cycles may yield erroneous results.

- If paired sera are to be collected, acute samples should be collected as soon as possible after the onset of symptoms. The second sample should be collected 14 to 21 days after the acute specimen was collected. Both samples must be run in duplicate on the same plate to test for a significant rise. If the first specimen is obtained late during the course of the infection, a significant rise may not be detectable.
- The NCCLS provides recommendations for storing blood specimens (Approved Standard - Procedures for the Handling and Processing of Blood Specimens, H18-A. 1990).<sup>18</sup>

### REAGENTS

#### Materials provided with the kit

Each kit contains the following components in sufficient quantities to perform the number of tests indicated on the package label.

- Toxoplasma antigen (inactivated) coated microassay plate:** 96 wells, configured in twelve 1x8 strips, stored in a foil pouch with desiccant. (one plate)
- Serum Diluent Type I:** Ready for use. Contains proclin (0.1%) as a preservative (one bottle, 30 mL)
- Calibrator:** Human serum or defibrinated plasma. Sodium azide (< 0.1%) and pen/strep (0.01%) added as preservatives, with kit specific factor printed on vial label. The Calibrator is used to calibrate the assay to account for day-to-day fluctuations in temperature and other testing conditions. (one vial, 0.4 mL)
- Positive Control:** Human serum or defibrinated plasma. Sodium azide (<0.1%) and pen/strep (0.01%) added as preservatives, with established range printed on vial label. The Positive Control is utilized to control the positive range of the assay. (one vial, 0.4 mL)\*
- Negative Control:** Human serum or defibrinated plasma. Sodium azide (<0.1%) and pen/strep (0.01%) added as preservatives, with established range printed on vial label. The Negative Control is utilized to control the negative range of the assay. (one vial, 0.4 mL) \*
- Horseradish-peroxidase (HRP) Conjugate:** Ready to use. Goat anti-human IgG, containing proclin (0.1%) and gentamicin as preservatives. (one bottle, 16 mL)
- Chromogen/Substrate Solution Type I:** Tetramethylbenzidine (TMB), ready to use. The reagent should remain closed when not in use. If allowed to evaporate, a precipitate may form in the reagent wells. (one bottle, 15 mL)
- Wash Buffer Type I (20X concentrate):** Dilute 1 part concentrate + 19 parts deionized or distilled water. Contains TBS, Tween-20 and proclin (0.1%) as a preservative. (one bottle, 50 mL)
- Stop Solution:** Ready to use, contains a 1N H<sub>2</sub>SO<sub>4</sub> solution. (one bottle, 15 mL)

\* Note: serum vials may contain excess volume

#### Materials required but not provided

- Wash bottle, automated or semi-automated microwell plate washing system.
- Micropipettes, including multichannel, capable of accurately delivering 10-200 µL volumes (less than 3% CV).
- One liter graduated cylinder.
- Paper towels.
- Test tube for serum dilution.
- Reagent reservoirs for multichannel pipettes.
- Pipette tips.

- Distilled or deionized water (dH<sub>2</sub>O), CAP (College of American Pathology) Type 1 or equivalent.<sup>20, 21</sup>
- Timer capable of measuring to an accuracy of +/- 1 second (0 - 60 minutes).
- Disposal basins and 0.5% sodium hypochlorite (50 mL bleach in 950 mL dH<sub>2</sub>O).
- Single or dual wavelength microplate reader with 450 nm filter. If dual wavelength is used set the reference filter to 600-650 nm. Read the Operator's Manual or contact the instrument manufacturer to establish linearity performance specifications of the reader.

Note: Use only clean, dry glassware.

### REAGENT PREPARATION

- All reagents must be removed from refrigeration and allowed to come to room temperature before use (21° to 25°C). Return all reagents to refrigerator promptly after use.
- All samples and controls should be vortexed before use.
- Dilute 50 mL of the 20X Wash Buffer Type I to 1 L with distilled and/or deionized H<sub>2</sub>O. Mix well.

### ASSAY PROCEDURE

Note: To evaluate paired sera, both serum samples must be tested in duplicate and run in the same plate. It is recommended that the serum pairs be run in adjacent wells.

- Place the desired number of strips into a microwell frame. Allow four (4) Control/Calibrator determinations (one Negative Control, two Calibrators and one Positive Control) per run. A reagent blank (RB) should be run on each assay. Check software and reader requirements for the correct Control/Calibrator configuration. Return unused strips to the sealable bag with desiccant, seal and immediately refrigerate.

#### Example Configuration:

Plate Location	Sample Description	Plate Location	Sample Description
1A	RB	2A	Patient #4
1B	NC	2B	Patient #5
1C	Cal	2C	Patient #6
1D	Cal	2D	Patient #7
1E	PC	2E	Patient #8 (Acute 1)
1F	Patient #1	2F	Patient #8 (Acute 2)
1G	Patient #2	2G	Patient #8 (Convalescent 1)
1H	Patient #3	2H	Patient #8 (Convalescent 2)

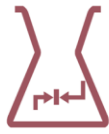
RB = Reagent Blank - Well without serum addition run with all reagents. Utilized to blank reader.

NC = Negative Control

Cal = Calibrator

PC = Positive Control

- Dilute test sera, Calibrator and Control sera 1:21 (e.g., 10 µL + 200 µL) in Serum Diluent. Mix well. (For manual dilutions it is suggested to dispense the Serum Diluent into the test tube first and then add the patient serum.)
- To individual wells, add 100 µL of the appropriate diluted Calibrator, Controls and patient sera. Add 100 µL of Serum Diluent to reagent blank well. Check software and reader requirements for the correct reagent blank well configuration.
- Incubate each well at room temperature (21 to 25°C) for 25 minutes +/- 5 minutes.



- Aspirate or shake out liquid from all wells. If using semi-automated or automated washing equipment add 250-300 µL of diluted Wash Buffer to each well. Aspirate or shake out to remove all liquid. Repeat the wash procedure two times (for a total of three (3) washes) for manual or semi-automated equipment or four times (for a total of five (5) washes) for automated equipment. After the final wash, blot the plate on paper toweling to remove all liquid from the wells.

**\*\*IMPORTANT NOTE: Regarding steps 5 and 8 - Insufficient or excessive washing will result in assay variation and will affect validity of results. Therefore, for best results the use of semi-automated or automated equipment set to deliver a volume to completely fill each well (250-300 µL) is recommended. A total of up to five (5) washes may be necessary with automated equipment. Complete removal of the Wash Buffer after the last wash is critical for the accurate performance of the test. Also, visually ensure that no bubbles are remaining in the wells.**

- Add 100 µL Conjugate to each well, including reagent blank well. Avoid bubbles upon addition as they may yield erroneous results.
- Incubate each well at room temperature (21 to 25°C) for **25 minutes +/- 5 minutes.**
- Repeat wash as described in Step 5.
- Add 100 µL Chromogen/Substrate Solution (TMB) to each well, including the reagent blank well, maintaining a constant rate of addition across the plate.
- Incubate each well at room temperature (21 to 25°C) for **10-15 minutes.**
- Stop reaction by addition of 100 µL of Stop Solution (1N H<sub>2</sub>SO<sub>4</sub>) following the same order of Chromogen/Substrate addition, including the reagent blank well. Tap the plate gently along the outsides, to mix contents of the wells. The plate may be held up to 1 hour after addition of the Stop Solution before reading.
- The developed color should be read on an ELISA plate reader equipped with a 450 nm filter. If dual wavelength is used, set the reference filter to 600-650 nm. The instrument should be blanked on air. The reagent blank must be less than 0.150 Absorbance at 450 nm. If the reagent blank is  $\geq 0.150$  the run must be repeated. Blank the reader on the reagent blank well and then continue to read the entire plate. Dispose of used plates after readings have been obtained.

### RESULTS

#### CALCULATIONS

- Mean Calibrator O.D. (Optical Density) - Calculate the mean O.D. value from the two Calibrator determinations.
- Correction Factor - To account for day-to-day fluctuations in assay activity due to room temperature and timing, a Correction Factor is determined by Diagnostic Automation/Cortez Diagnostics, Inc. for each lot of kits. The Correction Factor is printed on the Calibrator vial.
- Cutoff Calibrator Value - The Cutoff Calibrator Value for each assay is determined by multiplying the Correction Factor by the mean Calibrator O.D. determined in Step 1.
- ISR Value - Calculate an Immune Status Ratio (ISR) for each specimen by dividing the specimen O.D. Value by the Cutoff Calibrator Value determined in Step 3.

Example: O.D.s obtained for Calibrator = 0.38, 0.42  
 Mean O.D. for Calibrator = 0.40  
 Correction Factor = 0.50  
 Cutoff Calibrator Value =  $0.50 \times 0.40 = 0.20$   
 O.D. obtained for patient sera = 0.60  
 ISR Value =  $0.60/0.20 = 3.00$

#### ANALYSIS

- The patients' ISR (Immune Status Ratio) values are interpreted as follows:

ISR	Results	Interpretation
$\leq 0.90$	Negative	No detectable antibody to Toxoplasma by the ELISA test. Such individuals are presumed to be uninfected with Toxoplasma and to be susceptible to primary infection.
0.91 – 1.09	Equivocal	Samples should be retested. See Number (3) below.
$\geq 1.10$	Positive	Indicates presence of detectable antibody to Toxoplasma by the ELISA test. Indicative of current or previous infection. The individual may be at risk of transmitting Toxoplasma infection, but is not necessarily currently contagious.

- Samples that remain equivocal after repeat testing should be retested on an alternate method, e.g., immunofluorescence assay (IFA). If results remain equivocal upon further testing, an additional sample should be taken. (See Limitation No. 4).
- In the evaluation of paired sera, if the acute specimen is negative and the convalescent specimen is positive, a seroconversion has taken place. This indicates a significant change in antibody level and the patient is undergoing a primary infection.
- To evaluate paired sera for a significant change in antibody level or seroconversion, both samples must be tested in duplicate in the same assay. The mean ISR of both samples (acute and convalescent) must be greater than 1.00 to evaluate the paired sera for significant rise in antibody level.
- Additional Quality Control for Paired Sera: (See NOTE under Assay Procedure). As a check for acceptable reproducibility of both the acute sera (tested in duplicate) and the convalescent sera (tested in duplicate), the following criteria must be met for valid results:

Acute 1 ISR = 0.8 to 1.2                      Convalescent 1 ISR= 0.8 to 1.2  
 Acute 2 SR    Convalescent 2 SR

- Compare the ISR of the pairs by calculating as follows:

$\frac{\text{Mean ISR (second sample)} - \text{Mean ISR (first sample)}}{\text{Mean ISR (first sample)}} \times 100 = \% \text{ RISE IN ISR LEVEL}$

#### INTERNATIONAL UNIT CONVERSION

International unit (IU) reactivity is determined relative to the IU standard. Conversion of Index values to international units is accomplished by using an exponential regression analysis. Each lot is standardized versus international units and provided with a lot specific conversion table (Conversion of International Units (IU) per mL for Toxoplasma IgG). For example:

ISR	IU
1.0	1.08
1.5	2.08
2.0	4.03
2.5	7.79

See attached addendum for the lot specific conversion table. Values were determined using the WHO International Standard Anti-Toxoplasma IgG, Human NIBSC code 01/600 which contains 20 IU per mL.



% Rise in ISR	Interpretation
<30.0 %	No significant change in antibody level. No evidence of recent infection. If active disease is still suspected, a third sample should be collected and tested in the same assay as the first sample to look for a significant rise in antibody level.
≥ 30.0%	Statistically significant change in antibody level detected. This identifies those persons who are presumed to be experiencing recent or current episodes of Toxo infection (reactivation, reinfection or a primary infection where the acute specimen was obtained too late to demonstrate seroconversion).

*Note: When evaluating paired sera, it should be determined if samples with high absorbance values are within linearity specifications of the spectrophotometer. Read the Operator's Manual or contact the instrument's manufacturer to obtain the established linearity specifications of your spectrophotometer.*

### QUALITY CONTROL

For the assay to be considered valid the following conditions must be met:

1. Calibrator and Controls must be run with each test run.
2. Reagent blank (when read against air blank) must be < 0.150 Absorbance (A) at 450 nm.
3. Negative Control must be ≤ 0.250 A at 450 nm (when read against reagent blank).
4. Each Calibrator must be ≥ 0.250 A at 450 nm (when read against reagent blank).
5. Positive Control must be ≥ 0.500 A at 450 nm (when read against reagent blank).
6. The ISR (Immune Status Radio) Values for the Positive and Negative Control should be in their respective ranges printed on the vials. If the Control values are not within their respective ranges, the test should be considered invalid and should be repeated.
7. Additional Controls may be tested according to guidelines, or requirements of local, state, and/or federal regulations or accrediting organizations
8. Refer to NCCLS C24-A for guidance on appropriate QC practices.<sup>19</sup>
9. If above criteria are not met upon repeat testing, contact Diagnostic Automation, Inc. Technical Services.

### PERFORMANCE CHARACTERISTICS

#### SENSITIVITY AND SPECIFICITY

A total of 106 samples from a random population were tested by the Diagnostic Automation, Inc. Toxoplasma IgG ELISA kit and a commercially available Toxoplasma IFA kit. Complete agreement was found for 101 of the samples, of which 63 (62%) were negative and 38 (38%) were positive. Five samples were found negative by the Diagnostic Automation, Inc. ELISA and positive (1:16, 1:16, 1:32, 1:32, and 1:32) by IFA. This data indicates 95.3% sensitivity and 100% specificity when the Diagnostic Automation, Inc. Toxoplasma IgG ELISA is compared to the IFA technique. Upon further testing with an alternate method as referee, all five samples were found to be negative by consensus.

Forty-seven of the 106 samples were also compared with another commercially available Toxoplasma IgG ELISA kit. Complete agreement was found for twenty-two samples that were positive and twenty-four samples that were negative. One sample tested positive on the Diagnostic Automation,

Inc. ELISA kit and negative on the other ELISA kit. After further testing, with the commercially available Toxoplasma IgG IFA kit used as a referee method, the sample was confirmed positive.

Ten paired sera (twenty samples), were evaluated with the Diagnostic Automation, Inc. Toxoplasma IgG ELISA kit, another commercially available ELISA kit and a commercially available Toxoplasma IFA kit. Each pair was taken from an individual with diagnosed Toxoplasma infection. Six of the individuals were positive for seroconversion (negative on acute and positive on convalescent) and four of the individuals were positive on both acute and convalescent samples. All ten pairs showed a significant rise in antibody level. Complete agreement, 100% sensitivity was established for the pairs when compared to the other ELISA test and the IFA.

#### REPRODUCIBILITY

Three studies were performed to assess the precision of the TMB test results. Five sera were used in 20 wells each ranging from negative to high positive for the inter-run assay. Five sera were used for 5 days in 5 wells each ranging from negative to high positive for the inter-day assay. Five sera were used in 3 wells each ranging from negative to high positive with 3 different lots of TMB Substrate. The summary of results are as follows:

Table 1

#### Intra-Run Assay

	Serum 1	Serum 2	Serum 3	Serum 4	Serum 5
Mean =	1.425	4.275	3.239	0.318	0.182
S.D. =	0.107	0.087	0.187	0.023	0.016
C.V. =	7.5%	2.0%	5.8%	7.2%	8.7%

Table 2

#### Inter-Day

	Serum 1	Serum 2	Serum 3	Serum 4	Serum 5
Mean =	1.644	5.632	4.223	0.406	0.286
S.D. =	0.191	0.414	0.273	0.058	0.015
C.V. =	11.6%	7.4%	6.5%	14.4%	5.4%

Table 3

#### Inter-Lot Assay

	Serum 1	Serum 2	Serum 3	Serum 4	Serum 5
Mean =	1.81	6.40	4.565	0.39	0.266
S.D. =	0.22	0.347	0.322	0.063	0.062
C.V. =	12.1%	5.4%	7.0%	16.2%	23.3%

#### INTERNATIONAL UNIT CONVERSION

The data in Table 4 illustrate Toxoplasma IgG ISR Values for the serially diluted international unit standard, obtained from the World Health Organization. The Toxoplasma IgG ISR Values are compared to serial dilutions of the international unit standard serum by linear regression (exponential regression analysis). The data indicate that international units can be determined from the ISR Value.

Table 4

#### International Unit Conversion\*

International Unit Standard Units / mL	ISR Value
19.61	3.2
15.07	3.0
7.79	2.5
4.03	2.0
1.08	1.0
0.83	0.8



Linear regression compared ISR Values versus International Units.

$$r^2 = 0.983 \quad a = 0.758 \quad b = 0.944 \quad Y = \text{ISR} \quad X = \text{IU} / \text{mL}$$

Exponential Regression Equation Calculation

$$X = \frac{(y+b)}{a} \quad e^x = \text{derived IU} / \text{mL}$$

Each lot is standardized versus international units and provided with a lot specific conversion table (Conversion of International Units (IU) per mL for Toxoplasma IgG).

\*Values were determined using the WHO International Standard Anti-Toxoplasma IgG, Human NIBSC code 01/600 which contains 20 IU per mL.

### LIMITATIONS OF THE ASSAY

1. Use fresh serum samples or samples frozen only once and thawed at 37°C. Samples that are improperly stored or are subjected to multiple freeze-thaw cycles may yield spurious results.
2. The user of this kit is advised to carefully read and understand the package insert. Strict adherence to the protocol is necessary to obtain reliable test results. In particular, correct sample and reagent pipetting, along with careful washing and timing of the incubation steps are essential for accurate results.
3. This kit is designed to measure IgG antibody in patient samples. Positive results in neonates must be interpreted with caution, since maternal IgG is transferred passively from the mother to the fetus before birth. IgM assays are generally more useful indicators of infection in children below 6 months of age.
4. Samples collected very early in the course of an infection may not have detectable levels of IgG. In such cases, it is recommended that an IgM assay be performed, or a second serum sample be obtained at a later date to be tested in parallel with the original sample to determine seroconversion.<sup>15, 16</sup>
5. The results of ELISA performed on serum from patients with immunosuppression must be interpreted with caution. The presence of IgG antibody against a particular virus or organism may not assure protection from that disease. For example, cases of reactivation of Toxoplasma infection in immunocompromised individuals have been documented.<sup>17</sup> Alternatively, certain immune individuals have been shown to have such low circulating IgG levels that they may appear negative for that antibody when tested.<sup>6</sup>
6. Samples that remain equivocal after repeat testing should be retested on an alternate method, e. g., immunofluorescence assay (IFA). If results remain equivocal upon further testing, an additional sample should be taken (See Limitation No. 4).
7. The values obtained from this assay are intended to be an aid to diagnosis only. Each physician must interpret the results in light of the patient's history, physical findings and other diagnostic procedures.

### STORAGE AND STABILITY

1. Store unopened kit between 2° and 8°C. The test kit may be used throughout the expiration date of the kit. Refer to the package label for the expiration date.
2. Unopened microassay plates must be stored between 2° and 8°C. Unused strips must be immediately resealed in a sealable bag with desiccant and returned to storage between 2° and 8°C.

3. Store HRP Conjugate between 2° and 8°C.
4. Store the Calibrator, Positive and Negative Controls between 2° and 8°C.
5. Store Serum Diluent Type I and 20X Wash Buffer Type 1 between 2° and 8°C.
6. Store the Chromogen/Substrate Solutions Type 1 between 2° and 8°C. The reagent should remain closed when not in use. If allowed to evaporate, a precipitate may form in the reagent wells.
7. Store 1X (dilute) Wash Buffer Type 1 at room temperature (21° to 25°C) for up to 5 days or up to 1 week between 2° and 8°C.

*Note: If constant storage temperature is maintained, reagents and substrate will be stable for the dating period of the kit. Refer to package label for expiration date. Precautions were taken in the manufacture of this product to protect the reagents from contamination and bacteriostatic agents have been added to the liquid reagents. Care should be exercised to protect the reagents in this kit from contamination.*

### PRECAUTIONS

1. For *in vitro* diagnostic use.
2. The human serum components used in the preparation of the Controls and Calibrator in this kit have been tested by an FDA approved method for the presence of antibodies to human immunodeficiency virus1 & 2 (HIV 1&2), hepatitis C (HCV) as well as hepatitis B surface antigen and found negative. Because no test method can offer complete assurance that HIV, HCV, hepatitis B virus, or other infectious agents are absent, specimens and human-based reagents should be handled as if capable of transmitting infectious agents.
3. The Centers for Disease Control & Prevention and the National Institutes of Health recommend that potentially infectious agents be handled at the Biosafety Level 2.<sup>15</sup>
4. The components in this kit have been quality control tested as a Master Lot unit. Do not mix components from different lot numbers except Chromogen/Substrate Solution Type I, Stop Solution, Wash Buffer Type I. Do not mix with components from other manufacturers.
5. Do not use reagents beyond the stated expiration date marked on the package label.
6. All reagents must be at room temperature (21° to 25°C) before running assay. Remove only the volume of reagents that is needed. **Do not pour reagents back into vials as reagent contamination may occur.**
7. Before opening Control and Calibrator vials, tap firmly on the benchtop to ensure that all liquid is at the bottom of the vial.
8. Use only distilled or deionized water and clean glassware.
9. Do not let wells dry during assay; add reagents immediately after completing wash steps.
10. Avoid cross-contamination of reagents. Wash hands before and after handling reagents. **Cross-contamination of reagents and/or samples could cause false results.**
11. If washing steps are performed manually, wells are to be washed three times. Up to five wash cycles may be necessary if a washing manifold or automated equipment is used.
12. **Sodium azide inhibits Conjugate activity. Clean pipette tips must be used for the Conjugate addition so that sodium azide is not carried over from other reagents.**
13. It has been reported that sodium azide may react with lead and copper in plumbing to form explosive compounds. When disposing, flush drains with water to minimize build-up of metal azide compounds.
14. Never pipette by mouth or allow reagents or patient sample to come into contact with skin. Reagents containing ProClin®, sodium azide, and TMB may be irritating. Avoid contact with skin and eyes. In case of contact, flush with plenty of water.



15. If a sodium hypochlorite (bleach) solution is being used as a disinfectant, do not expose to work area during actual test procedure because of potential interference with enzyme activity.
16. Avoid contact of Stop Solution (1N sulfuric acid) with skin or eyes. If contact occurs, immediately flush area with water.
17. **Caution:** Liquid waste at acid pH must be neutralized prior to adding sodium hypochlorite (bleach) solution to avoid formation of poisonous gas. Recommend disposing of reacted, stopped plates in biohazard bags. See Precaution 3.
18. The concentrations of anti-Toxoplasma in a given specimen determined with assays from different manufacturers can vary due to differences in assay methods and reagent specificity.

The safety data sheet is available upon request.



**WARNING**

*Serum Diluent, Conjugate, and Wash Buffer contain 0.1% ProClin 300R, a biocidal preservative that may cause sensitization by skin contact; prolonged or repeated exposure may cause allergic reaction in certain sensitive individuals.*  
**H317: May cause an allergic skin reaction.**

**P280:** Wear protective gloves / protective clothing / eye protection / face protection.

**P302 + P352:** IF ON SKIN: Wash with plenty of soap and water.

**P333 + P313:** If skin irritation or rash occurs: Get medical advice/ attention.

**P501:** Dispose of contents and container in accordance to local, regional, national and international regulations.

**WARNING**

*Serum Diluent and Controls contain < 0.1% sodium azide.*

**H302:** Harmful if swallowed

**P264:** Wash thoroughly with plenty of soap and water after handling

**P270:** Do not eat, drink or smoke when using this product

**P301+P312:** IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell

**P330:** If swallowed, rinse mouth

**P501:** Dispose of contents/container to in accordance to local, regional, national and international regulations.

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**MANUFACTURER AND BRAND DETAILS**

**ISO 13485:2016**

ISO 13485  
Quality  
Management for  
Medical Devices  
CERTIFIED

**Diagnostic Automation/Cortez Diagnostics, Inc.**  
 21250 Califa Street, Suite 102 and 116,  
 Woodland Hills, California 91367 USA

<b>Date Adopted</b>	2023-11
<b>Brand Name</b>	AccuDiag™
<b>REF</b> 1101	AccuDiag™ - <i>Toxoplasma gondii</i> IgG (Toxo IgG) ELISA
<b>PIC</b>	PB1101ZY1
<b>Revision Date: 2022-04</b>	



## SUMMARY OF ASSAY PROCEDURE

DILUTE  
SERUM

