Snibe Diagnostic

CE 0123 REF 130212002M: 100 tests
130612002M: 50 tests

MAGLUMI® Toxo IgM (CLIA)

INTENDED USE

The kit is an *in vitro* chemiluminescence immunoassay for the qualitative determination of Toxo IgM in human serum to aid in the diagnosis of acute or recent Toxoplasma gondii infection using the MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer (including Maglumi 600, Maglumi 800, Maglumi 1000, Maglumi 1000 Plus, Maglumi 2000, Maglumi 2000 Plus, Maglumi 4000, Maglumi 4000 Plus and MAGLUMI X8).

SUMMARY AND EXPLANATION OF THE TEST

Toxoplasmosis is a parasitic disease caused by Toxoplasma gondii[1]. Toxoplasma gondii was first described in 1908 by Nicolle and Manceaux in Tunisia, and independently by Splendore in Brazil.[2] Splendore reported the protozoan in a rabbit, while Nicolle and Manceaux identified it in a North African rodent, the gundi (Ctenodactylus gundi)[3]. In 1909 Nicolle and Manceaux differentiated the protozoan from Leishmania[2]. Nicolle and Manceaux then named it Toxoplasma gondii after the curved shape of its infectious stage (Greek root 'toxon'= bow)[2].

Infections with toxoplasmosis usually cause no obvious symptoms in adults[4]. Occasionally there may be a few weeks or months of mild flu-like illness such as muscle aches and tenderlymph nodes[5]. In a small number of people, eye problems may develop[5]. In those with a weak immune system, severe symptoms such as seizures and poor coordination may occur[5]. Toxoplasmosis is usually spread by eating poorly cooked food that contains cysts, exposure to infected cat feces, and from a mother to a child during pregnancy if the mother becomes infected[1]. Rarely the disease may be spread by blood transfusion[1]. It is not otherwise spread between people[1]. The parasite is only known to reproduce sexually in the cat family[7]. However, it can infect most types of warm-blooded animals, including humans[7]. Diagnosis is typically by testing blood for antibodies or by testing amniotic fluid for the parasite's DNA[6].

Transplacental transmission of the parasite is possible and congenital infections occur in about 45% of pregnant women with acute phase toxoplasmosis. The severity of the congenital abnormalities is higher the earlier in the pregnancy the infection is acquired, but as the pregnancy progresses the risk of fetal infection increases[8]. Approximately 90% of neonates infected in utero are asymptomatic at birth but adverse sequelae, including chorioretinitis, convulsions, mental and psychomotor retardation may occur later in life[9].

IgG antibodies to Toxoplasma gondii usually appear within a week or two of infection, peak within one to two months, then decline at various rates[10]. In contrast to IgG, IgM antibodies can be used to detect acute infection, but generally not chronic infection. The IgM antibodies appear sooner after infection than the IgG antibodies and disappear faster than IgG antibodies after recovery. In most cases, T. gondii-specific IgM antibodies can first be detected approximately a week after acquiring primary infection, and decrease within one to six months; 25% of those infected are negative for T. gondii-specific IgM within seven months[11-12].

The determination of Toxo IgM antibodies, can aid in the diagnosis of diseases caused by Toxoplasma gondii, is used to assess the serological status of an individual and is indicative for an acute or past infection, together with the detection of Toxo IgG. This is particularly important in order to adopt suitable prophylaxis in susceptible individuals.

PRINCIPLE OF THE TEST

The Toxo IgM assay is an indirect chemiluminescence immunoassay.

The sample (or calibrator/control, if applicable), buffer (including goat Anti-human IgG, goat Anti-human IgA), magnetic microbeads coated with purified Toxo antigen are mixed thoroughly and incubated, forming antibody-antigen complexes. After precipitation in a magnetic field, decant the supernatant, and perform a wash cycle. Then add ABEI labeled with mouse anti-human IgM antibody, and incubate to form sandwich complexes. After precipitation in a magnetic field, decant the supernatant, and then perform another wash cycle. Subsequently, the Starter 1+2 are added to initiate a chemiluminescent reaction. The light signal is measured by a photomultiplier as relative light units (RLUs), which is indicative of the concentration of Toxo IgM present in the sample (or calibrator/control, if applicable).

KIT COMPONENTS

Material Provided

Components	Contents	100 tests (REF: 130212002M)	50 tests (REF: 130612002M)
Magnetic Microbeads	Magnetic microbeads coated with Toxo antigen, containing BSA, NaN_3 (<0.1%).	2.5 mL	2.0 mL
Calibrator Low	Toxo IgM, containing bovine serum, NaN_3 (<0.1%).	2.5 mL	2.0 mL
Calibrator High	Toxo IgM, containing bovine serum, NaN_3 (<0.1%).	2.5 mL	2.0 mL
Buffer	Goat anti-human IgA, goat anti-human IgG, containing BSA, NaN_3 (<0.1%).	25.0 mL	13.5 mL
ABEI Label	Mouse anti-human IgM labeled with ABEI, containing BSA, NaN_3 (<0.1%).	22.5 mL	12.5mL
Internal Quality Control	Toxo IgM, containing bovine serum, NaN_3 (<0.1%).	2.0 mL	2.0 mL
All reagents are provided ready-to-use.			

Accessories Required But Not Provided

MAGLUMI Series:

Reaction Module	REF: 630003
Starter 1+2	REF: 130299004M, 130299027M
Wash Concentrate	REF: 130299005M
Light Check	REF: 130299006M
Reaction Cup	REF: 130105000101

Please order accessories from Shenzhen New Industries Biomedical Engineering Co., Ltd. (SNIBE) or our authorized representatives.

CALIBRATION

Traceability: This method has been standardized against the SNIBE internal reference substance.

Test of assay specific calibrators allows the RLU values to adjust the assigned master curve. Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve (10 calibrations) provided via the reagent Radio Frequency Identification (RFID) CHIP.

Recalibration is recommended if any of the following conditions occurs:

- After each change of lots (Reagent or Starter 1+2).
- Every 4 weeks and/or each time a new reagent kit is used (recommended).
- After instrument service is required.
- If controls lie outside the expected range.

QUALITY CONTROL

Follow government regulations or accreditation requirements for quality control frequency.

Internal quality control is only applicable with MAGLUMI system. For instructions for use and target value refer to ***Toxo IgM (CLIA) Quality Control Information***. User needs to judge results with their own standards and knowledge.

For detailed information about entering quality control values, refer to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

To monitor system performance and chart trends, commercially available quality control materials are required. Treat all quality control samples the same as patient samples. A satisfactory level of performance is achieved when analyte values obtained are within the acceptable Control Range for the system or within your range, as determined by an appropriate internal laboratory quality control scheme. If the quality control results do not fall within the Expected Values or within the laboratory's established values, do not report results. Take the following actions:

- Verify that the materials are not expired.
- Verify that required maintenance was performed.
- Verify that the assay was performed according to the instructions for use.
- Rerun the assay with fresh quality control samples.
- If necessary, contact your local technical supporters or distributors for assistance.

SPECIMEN COLLECTION AND PREPARATION

- Use standard sampling tubes or tubes containing separating gel. Collect blood aseptically following the universal precautions for venipuncture.
- Ensure that complete clot formation in specimens has taken place prior to centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy, may exhibit increased clotting time.
- If the specimen is centrifuged before a complete clotting, the presence of fibrin may cause erroneous results. Samples must be free of fibrin and other particulate matter.
- Do not use hemolyzed or grossly lipemic specimens as well as specimens containing particulate substance or exhibiting obvious microbial contamination. Inspect all specimens for bubbles, and remove bubbles before analysis for optimal results.
- Avoid repeated freezing and thawing. The serum sample can be frozen and thawed for only two times. Stored samples should be thoroughly mixed prior to use (Vortex mixer). Frozen specimens must be mixed THOROUGHLY after thawing by LOW speed vortexing. Please ask local representative of SNIBE for more derails if you have any doubt.
- Centrifuged specimens with a lipid layer on the top must be transferred to a sample cup or a secondary tube. Care should be taken to transfer only the clarified specimen without the lipaemic material.
- All samples (patient specimens and controls) should be tested within 3 hours when placed on board the MAGLUMI System. Refer to the SNIBE service for more details of onboard sample storage constraints.
- Specimens removed from the separator, red blood cells or clot may be stored up to 7 days at 2-8°C, and stored up to 3 months frozen at -20°C or colder.
- Before shipping specimens, it is recommended that specimens be removed from the clot, red blood cells, or separator. When shipped, specimens should be packaged and labeled in compliance with applicable state, federal and international regulations covering the transport of clinical specimens and infectious substances. Specimens should be shipped frozen.
- The sample volume required for a single determination of Toxo IgM is 10 µL.

WARNING AND PRECAUTIONS FOR USERS

- **IVD**
- For *In Vitro* Diagnostic Use.
- Follow the package insert carefully. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.

Safety Precautions

- **CAUTION:** This product requires the handling of human specimens. It is recommended that all human sourced materials be considered potentially infectious and handled in accordance with the 29 CFR 1910.1030 Occupational exposure to bloodborne pathogens. Biosafety Level 2 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.
- All samples, biological reagents and materials used in the assay should be considered potentially able to transmit infectious agents. They should therefore be disposed in accordance with the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with prevailing regulatory requirements.
- This product contains Sodium Azide. Dispose of contents and containers must be in accordance with all local, regional and national regulations.
- Refer to safety data sheets which are available on request.

Handling Precautions

- Do not use reagent kits beyond the expiration date.
- Do not interchange reagent components from different reagents or lots.
- Prior to loading the reagent kit on the system for the first time, the reagent kit requires mixing to re-suspend magnetic microbeads that have settled during shipment.
- For magnetic microbeads mixing instructions, refer to the Preparation of the Reagent section of this package insert.
- To avoid contamination, wear clean gloves when operating with a reagent kit and samples.
- Over time, residual liquids may dry on the septum surface. These are typically dried salts which have no effect on assay efficacy.
- For detailed discussion of handling precautions during system operation, refer to the SNIBE service information.

STORAGE AND STABILITY

- Sealed: Stored at 2-8°C until the expiration date.
- Opened at 2-8°C: Minimum stability is 4 weeks.
- On-board: Minimum stability is 4 weeks.

- To ensure the best kit performance, it is recommended to place opened kits in the refrigerator after the end of the intraday test work. It is still possible to keep on using the kit beyond the opened or on-board period if the controls are found within the expected ranges.
- Keep upright for storage to facilitate later proper resuspension of magnetic microbeads.
- Keep away from sunlight.

TEST PROCEDURE

Preparation of the Reagent

- Resuspension of the magnetic microbeads takes place automatically when the kit is loaded successfully, ensuring the magnetic microbeads are totally resuspended homogenous prior to use.
- To ensure proper test performance, strictly adhere to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer. Each test parameter is identified via a RFID CHIP on the Reagent kit. For further information please refer to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

High-Dose Hook

Whenever samples containing extremely high antibody concentrations are tested, the saturation effect can mimic concentrations lower than real. However, a well-optimized two-step method excludes grossly underestimated results, because the analytical signals remain consistently high (saturation curve).

No false negative result due to high-dose hook effect was found with the Toxo IgM assay.

LIMITATIONS

- A skillful technique and strict adherence to the instructions are necessary to obtain reliable results.
- Bacterial contamination or heat inactivation of the specimens may affect the test results.
- A result within the expected range does not rule out the presence of disease and should be interpreted together with the patient's clinical picture and other diagnostic procedures.
- Diagnosis of a disease should not be based on the result of a single test, but should be determined in conjunction with clinical findings in association with medical judgement.
- Any therapeutical decision should also be taken on a case-by-case basis.
- Patient samples containing human anti-mouse antibodies (HAMA) may give falsely elevated or decreased values. Although HAMA-neutralizing agents are added, extremely high HAMA serum concentrations may occasionally influence results.

RESULTS

Calculation of Results

The analyzer automatically calculates the concentration in each sample by means of a calibration curve which is generated by a 2-point calibration master curve procedure. The results are expressed in AU/mL. For further information please refer to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

Interpretation of Results

Results obtained with the Toxo IgM assay can be interpreted as follows:

- Non-reactive: A result less than 2 AU/mL (<2 AU/mL) is considered to be negative. Individuals with such results are presumed to be not currently infected with Toxo.
- Gray zone: A result in the interval between 2 and 2.6 (2≤ x<2.6 AU/mL) is considered to be equivocal.
- Reactive: A result greater than or equal to 2.6 AU/mL is (≥2.6 AU/mL) considered to be positive. Reactivity for IgM antibodies to Toxo may indicate current infection, reactivation or recent vaccination.

NOTE:

- It is recommended to confirm results of specimens in gray zone by testing Toxo IgG.
- Consider to take a second sample, if possible, within an appropriate period of time (e.g., two weeks) to confirm levels of IgM and IgG.

Since there is no international standard material for Toxo IgM yet, different IVD manufacturer have different traceability chain. Therefore results from assays of other manufacturers cannot be used interchangeably.

PERFORMANCE CHARACTERISTICS

Precision

Precision for the Toxo IgM assay was determined as described in the CLSI EP5-A2, 1 controls and 3 human serum pools containing different concentration of analyte were assayed in duplicate at two independent runs per day for 20 testing days. The results are summarized in the following table:

Sample	Mean(AU/mL) (N=80)	Within-Run		Between-Run		Total	
		SD(AU/mL)	%CV	SD(AU/mL)	%CV	SD(AU/mL)	%CV
Negative Serum Pool	1.508	0.061	4.05	0.085	5.64	0.105	6.96
Low Positive Serum Pool	6.519	0.280	4.30	0.201	3.08	0.344	5.28
High Positive Serum Pool	20.018	0.181	0.90	0.577	2.88	0.604	3.02
Positive Control	8.555	0.252	2.95	0.218	2.55	0.333	3.89

Analytical Sensitivity

<0.25 AU/mL

The Analytical Sensitivity represents the lowest analyte level that can be distinguished from zero.

Recovery

Consider calibrator high of known concentration as a sample, dilute it by 1:2 ratio with diluents, and measure its diluted concentration for 10 times. Then calculate the expected concentration and recovery of measured concentration. The recovery should be within 90% -110%.

Expected	Mean Measuring	%Recovery
9.800 AU/mL	9.724 AU/mL	99.22

Analytical Specificity

Clinical Toxo IgM negative samples, which contain potential cross-reactants including HAV, HBV, HCV, HIV, Syphilis, EBV, CMV IgM, Rubella IgM, Toxo IgG, HSV-1/2 IgM, RF, HAMA, ANA approved by commercially available CE-marked assay, were used to evaluate the cross-reactivity of Toxo IgM assay. Of all the potential cross-reactants, none were found to cause false positive in the Toxo IgM assay.

Endogenous Interference

Substances up to the following concentrations did not interfere with the assay:

- Bilirubin 40 mg/dL
- Hemoglobin 1000 mg/dL
- Triglyceride 2000 mg/dL

Drug Interference

Drugs up to the following concentrations did not interfere with the assay:

Drugs	Concentration
N-acetylcysteine	150 µg/mL
Methyldopa	25 µg/mL
Theophylline	60 µg/mL
Metformin	12 µg/mL
Isosorbide dinitrate	6 µg/mL
Rifampicin	48 µg/mL
Doxycycline	18 µg/mL
Cefoxitin	6600 µg/mL
Cyclosporine	2 µg/mL
Metronidazole	125 µg/mL
Ascorbic acid	60 µg/mL
Phenylbutazone	200 µg/mL
Aspirin	1000 µg/mL
Acetaminophen	400 µg/mL
Ibuprofen	500 µg/mL
Sodium salicylate	500 µg/mL

REFERENCES

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SYMBOLS EXPLANATIONS

	Consult instructions for use		Manufacturer
8 °C 2 °C	Temperature limit (Store at 2-8 °C)		Use-by date
Σ	Contains sufficient for		Keep away from sunlight
	This way up	EC REP	Authorized representative in the European Community
IVD	*In vitro* diagnostic medical device	CONTENTS	Kit components
REF	Catalogue number	LOT	Batch code