

GENLISA® Chicken Toxoplasma-Immunoglobulin M, Tox-IgM ELISA

REF : KLC0725

Ver 1.0

RUO

Enzyme Immunoassay for the Qualitative Determination of Toxoplasma-Immunoglobulin M, Tox-IgM in Chicken serum

RUO	For Research Use Only	REF	Catalog Number
	Store At	LOT	Batch Code
	Manufactured By		Biological Risk
	Expiry Date		Consult Operating Instructions

For Research Use Only. Purchase does not include or carry the right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of Krishgen Biosystems Private Limited is strictly prohibited.

REF KLC0725 **96 tests****Krishgen Biosystems Private Limited**

For US/Europe Customers: toll free +1(888)-970-0827 | tel +1(562)-568-5005

For Asia/India Customers: tel +91(22)-49198700

Email: sales1@krishgen.com | <http://www.krishgen.biz> / www.krishgenbio.com

Introduction:

The GENLISA® ELISA kits are used for assessing the specific biomarker in samples analytes which may be serum, plasma and cell culture supernatant as validated with the kit. The kit employs an indirect ELISA technique.

Intended Use:

The GENLISA® Chicken Toxoplasma-Immunoglobulin M, Tox-IgM ELISA kit is used as an analytical tool for qualitative determination of Chicken Toxoplasma-Immunoglobulin M, Tox-IgM in serum.

Principle:

The method employs indirect ELISA technique. Antigens are pre-coated onto microwells. Samples and controls are pipetted into microwells and Chicken Toxoplasma-Immunoglobulin M, Tox-IgM present in the sample are bound by the antigens. Anti-Chicken Antibody IgM conjugated to HRP is pipetted and incubated to form a complex. After washing microwells in order to remove any non-specific binding, the substrate solution (TMB) is added to microwells and color develops proportionally to the amount of Chicken Toxoplasma-Immunoglobulin M, Tox-IgM in the sample. Color development is then stopped by addition of stop solution. Absorbance is measured at 450 nm.

Materials Provided:

1. Chicken Tox-IgM Antibody Microtitre coated plate (8 x 12 wells) - 1 no.
2. Positive Control – 1 ml
3. Negative Control - 1 ml
4. Anti-Chicken Antibody IgM:HRP Conjugate – 12 ml
5. Sample Diluent – 12 ml
6. (20X) Wash Buffer - 25 ml
7. TMB Substrate - 12 ml
8. Stop Solution - 12 ml
9. Instruction manual

Materials to be provided by the End-User:

1. Microtiter Plate Reader able to measure absorbance at 450 nm.
2. Adjustable pipettes and multichannel pipettor to measure volumes ranging from 25 ul to 1000 ul.
3. Deionized (DI) water.
4. Wash bottle or automated microplate washer.
5. Clean tubes and Eppendorf tubes.
6. Precision single and multi-channel pipette and disposable tips.
7. 37°C incubator.
8. Timer.

Handling/Storage:

1. All reagents should be stored as indicated on the component label.
2. All the reagents and wash solutions should be used within 12 months from manufacturing date.
3. Before using, bring all components to room temperature (18-25°C). Upon assay completion ensure all components of the kit are returned to appropriate storage conditions.
4. The Substrate is light-sensitive and should be protected from direct sunlight or UV sources.

Health Hazard Warnings:

1. Reagents that contain preservatives may be harmful if ingested, inhaled or absorbed through the skin.
2. For Research Use Only.

Specimen Collection and Preparation:

Isolate the test samples soon after collecting, then, analyze immediately (within 2 hours). Or aliquot and store at -20°C for long term. Avoid multiple freeze-thaw cycles.

- Serum: Coagulate the serum at room temperature (about 1 hour). Centrifuge at approximately 1000 × g for 15 min. Analyze the serum immediately or aliquot and store at -20°C.

Note: Samples to be used within 5 days may be stored at 2-8°C, otherwise samples must be stored at -20°C (≤1 month) or -80°C (≤2 months) to avoid loss of bioactivity and contamination. Hemolyzed samples are not suitable for use in this assay.

Reagent Preparation (all reagents should be diluted immediately prior to use):

1. Label any aliquots made with the kit Lot No and Expiration date and store it at appropriate conditions mentioned.
2. Bring all reagents to Room temperature before use.
3. To make **Wash Buffer (1X)**; dilute **25ml** of **20X Wash Buffer** in **475 ml** of **DI water**.

Procedural Notes:

1. In order to achieve good assay reproducibility and sensitivity, proper washing of the plates to remove excess un-reacted reagents is essential.
2. High Dose Hook Effect may be observed in samples with very high concentrations of Chicken Toxoplasma-Immunoglobulin M, Tox-IgM. High Dose Hook Effect is due to excess of antibody for very high concentrations of Chicken Toxoplasma-Immunoglobulin M, Tox-IgM present in the sample. High Dose Hook effect is most likely encountered from samples early in the purification process. If Hook Effect is possible, the samples to be assayed should be diluted with a compatible diluent.
3. Chicken Toxoplasma-Immunoglobulin M, Tox-IgM concentration of the undiluted sample is less than the diluted sample, this may be indicative of the Hook Effect.
4. Avoid assay of Samples containing sodium azide (NaN₃), as it could destroy the HRP activity resulting in under-estimation of the amount of Chicken Toxoplasma-Immunoglobulin M, Tox-IgM.
5. It is recommended that all Controls and Samples be assayed in duplicates or triplicates.
6. Maintain a repetitive timing sequence from well to well for all the steps to ensure that the incubation timings are same for each well.
7. If the Substrate has a distinct blue color prior to use it may have been contaminated and use of such substrate can lead to compromisation of the sensitivity of the assay.
8. The plates should be read within 30 minutes after adding the Stop Solution.
9. Make a work list in order to identify the location of Controls and Samples.

Assay Procedure:

1. Bring all reagents to room temperature prior to use. It is strongly recommended that Samples should be run in duplicates.
2. Add **100 ul Negative control and Positive control** to respective control wells except blank wells.
3. Add **100 ul** of **Sample Diluent** to each of the other reaction wells and add **10 ul** of test specimen in order. The color change effect is better when the sample is added and mixed by blowing.
4. Cover the plate with the sealer and incubate for 30 minutes at 37°C.
5. Aspirate and wash plate 5 times with diluted Wash Buffer (1X) and blot residual buffer by firmly tapping plate upside down on absorbent paper. Wipe of any liquid from the bottom outside of the microtiter wells as any residue can interfere in the reading step.
6. Add **100 ul** of **Anti-Chicken Antibody IgM:HRP Conjugate** to each well except blank well. Mix well.
7. Cover the plate with the sealer and incubate for 30 minutes at 37°C.

8. Aspirate and wash as per step no. 5.
9. Pipette **100 ul TMB Substrate** in each well.
10. Incubate the plate at **37°C** for **15 minutes**. DO NOT SHAKE or else it may result in higher backgrounds and worse precision. Positive wells should turn bluish in color.
11. Pipette **100 ul of Stop Solution** to all wells. The wells should turn from blue to yellow in color.
12. Read the absorbance at 450 nm with a microplate within 10-15 minutes after addition of Stop solution.

Calculation of Results:

Cut Off Value (C.O.) = NCx + 0.10

NCx: Mean Absorbance of Negative Control.

Validity of the test:

Mean of the Positive Control ≥ 0.8

Mean of the Negative Control ≤ 0.1 (If one well OD>0.1, it should be abandoned; If two wells A>0.1, it should be retested)

Interpretation of Results

Negative determinant: if the sample OD value $\leq$ Cut Off, the Chicken Toxoplasma-Immunoglobulin M, Tox-IgM is Negative.

Positive determinant: if the sample OD value $>$ Cut Off, the Chicken Toxoplasma-Immunoglobulin M, Tox-IgM is Positive.

Safety Precautions:

- **This kit is For Research Use only.** Follow the working instructions carefully.
- The expiration dates stated on the kit are to be observed. The same relates to the stability stated for reagents.
- Do not use or mix reagents from different lots.
- Do not use reagents from other manufacturers.
- Avoid time shift during pipetting of reagents.
- All reagents should be kept in the original shipping container.
- Since the kit contains potentially hazardous materials, the following precautions should be observed.
 - Do not smoke, eat or drink while handling kit material.
 - Always use protective gloves.
 - Never pipette material by mouth.
 - Wipe up spills promptly, washing the affected surface thoroughly with a decontaminant.
- In any case GLP should be applied with all general and individual regulations to the use of this kit.



Typical Example of a Work List

Well #	Contents	Absorbance at 450nm	Mean Absorbance	Interpolated Concentration
1A	Blank			
2A	Blank			
1B	Positive Control			
2B	Positive Control			
1C	Negative Control			
2C	Negative Control			
1D	Sample			
2D	Sample			
1E	Sample			
2E	Sample			
1F	Sample			
2F	Sample			
1G	Sample			
2G	Sample			
1H	Sample			
2H	Sample			
3A	Sample			
4A	Sample			
3B	Sample			
4B	Sample			

LIMITED WARRANTY

Krishgen Biosystems Private Limited does not warrant against damages or defects arising in shipping or handling, or out of accident or improper or abnormal use of the Products; against defects in products or components not manufactured by Krishgen Biosystems Private Limited, or against damages resulting from such non-Krishgen Biosystems Private Limited made products or components. Krishgen Biosystems Private Limited passes on to customer the warranty it received (if any) from the maker thereof of such non Krishgen made products or components. This warranty also does not apply to Products to which changes or modifications have been made or attempted by persons other than pursuant to written authorization by Krishgen Biosystems Private Limited.

THIS WARRANTY IS EXCLUSIVE. The sole and exclusive obligation of Krishgen Biosystems Private Limited shall be to repair or replace the defective Products in the manner and for the period provided above. Krishgen Biosystems Private Limited shall not have any other obligation with respect to the Products or any part thereof, whether based on contract, tort, and strict liability or otherwise. Under no circumstances, whether based on this Limited Warranty or otherwise, shall Krishgen Biosystems Private Limited be liable for incidental, special, or consequential damages.

This Limited Warranty states the entire obligation of Krishgen Biosystems Private Limited with respect to the Products. If any part of this Limited Warranty is determined to be void or illegal, the remainder shall remain in full force and effect.

Krishgen Biosystems Private Limited. 2026

THANK YOU FOR USING A KRISHGEN PRODUCT!

KRISHGEN BIOSYSTEMS PRIVATE LIMITED®, DHARMAPLEX®, GENBULK®, GENLISA®, KRISHZYME®, KRISHGEN®, KRIBIOLISA®, KRISHPLEX®, TITANIUM®, QUALICHEK® are registered trademarks of KRISHGEN BIOSYSTEMS PRIVATE LIMITED.

©KRISHGEN BIOSYSTEMS PRIVATE LIMITED. ALL RIGHTS RESERVED.

KRISHGEN BIOSYSTEMS PRIVATE LIMITED | OUR REAGENTS | YOUR RESEARCH |

SYMBOLS KEY

	Coated Microtiter Plate (8 x 12 wells)
	Positive Control
	Negative Control
	HRP- Conjugate
	Sample Diluent
	(20X) Wash Buffer
	TMB Substrate
	Stop Solution
	Consult Instructions for Use
	Catalog Number
	Expiration Date
	Storage Temperature