

## Quantiplus® Toxoplasma Detection Kit (Real-Time Qualitative PCR Kit)



QL-TOXO-25 : 25 rxns  
 QL-TOXO-50 : 50 rxns  
 QL-TOXO-100 : 100 rxns

**RUO**  
 PI/QLTOXO-01

### Intended Use

Quantiplus® Toxoplasma Detection Kit is a Real-Time PCR based in vitro diagnostic assay for detection of Toxoplasma in Plasma, CSF, BAL fluids and biopsy tissues. The kit consists of Toxo Ready Mix, Toxo PC, and Internal Control (Huwel IC-B mix). The Huwel IC-B mix is a second amplification system used to identify possible PCR inhibition without affecting the analytical sensitivity of the assay.

### Background Information

Toxoplasma is a single-celled organism which causes Toxoplasmosis, the most common parasitic infection worldwide affecting animals as well as humans. This parasite can be carried by the vast majority of warm-blooded animals. The causative agent of toxoplasmosis, the disease, is usually minor and self-limiting but can have serious or even fatal effects on a fetus whose mother first contracts the disease during pregnancy. Acute stage toxoplasma infections can be asymptomatic, but often gives flu-like symptoms in the early acute stages, and like flu can become, in very rare cases, fatal. Latent infection is normally asymptomatic; however, in the case of immunocompromised patients (such as those infected with HIV or transplant recipients on immunosuppressive therapy), toxoplasmosis can develop. The most notable manifestation of toxoplasmosis in immunocompromised patients is toxoplasmic encephalitis, which can be deadly. If infection with Toxoplasma gondii occurs for the first time during pregnancy, the parasite can cross the placenta, possibly leading to hydrocephalus, intracranial calcification, and chorioretinitis, with the possibility of spontaneous abortion or intrauterine death.

### Kit Components

Color Coding (Caps)	Contents	Description	25 rxns (QL-TOXO-25)	50 rxns (QL-TOXO-50)	100 rxns (QL-TOXO-100)
<b>Amber</b>	Toxo Ready Mix	Tox, internal control probes, primers with amplification Mix	1 x 375 µL	1 x 750 µL	2 x 750 µL
<b>Natural</b>	Huwel IC-B Mix	Internal Control	1 x 300 µL	1 x 600 µL	2 x 600 µL
<b>Red</b>	Toxo PC	Toxoplasma positive control	1 x 100 µL	1 x 100 µL	2 x 100 µL
White	Huwel PW	Purified water	1 x 500 µL	1 x 500 µL	2 x 500 µL

**Note: Please pay attention to the cap color coding and the tube contents.**

**Huwel PW (Molecular biology grade purified water)**

### Storage and Transportation Conditions

The kit should be transported at temperature below -20 °C. The kit is stable until the expiry date mentioned on the package, if the storage temperature is within -20 ±5 °C. More than 4X freezing and thawing cycles reduces the assay sensitivity. For intermittent usage the reagents should be frozen in aliquots.

### Technical specifications

Target Sequence	Glycerol-3-phosphate dehydrogenase gene (B1 gene)
Specificity	100%
Sensitivity	0.2 copies/µL and 50 copies/ mL
Validated Specimen	Plasma (K2EDTA-Blood)

**Assay Procedure**

**DNA Extraction**

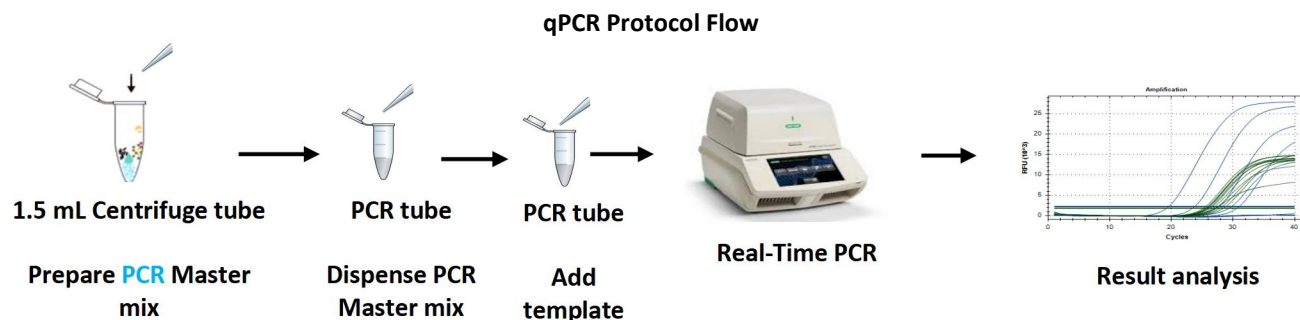
Quantiplus® Toxoplasma Detection Kit (Real-Time Qualitative PCR Kit) has been validated using the following DNA extraction kits. Recommended sample volume for extraction and elution are as follows:

S. No.	Name of the Extraction Kit	Recommended Sample volume for Extraction	Recommended Final Elution volume
1.	Huwel Nucleic Acid Extraction Kit - Version 2.0 (Cat. No. HL-NAX-100)	200 µL	100 µL
2.	QIAamp® DNA Blood Mini Kit (Cat. No. 51104)	200 µL	100 µL

*Note: Customer can also validate their own extraction process using other Viral DNA extraction Kits.*

*IC-B mix can be added at the extraction step or while setting up the PCR*

*The recommended sample volumes for extraction and elution are also applicable for CSF and BAL fluid sample types.*



**Preparation of Reaction Master mix**

Components	Volume per reaction (for 26µL)
Toxo Ready Mix	15.0
Huwel IC-B Mix (if not added at extraction step)	1.0
Extracted DNA/ Toxo PC / Huwel PW	10.0

It is necessary to keep all components at +2 °C to +8 °C during the PCR preparation. Close the tubes and centrifuge briefly before proceeding to the thermal cycler.

**Cycling Conditions**

Steps	No. of cycles	Temperature (°C)	Time
1 (Initial denaturation)	1	95	15 min.
2 (PCR cycling)	40	95	15 sec.
		60*	1 min.

**\*Plate read/Data acquisition in FAM and Yakima Yellow/ VIC/HEX channels**

**Sample analysis and Interpretation**

Interpret the test results as shown in the table below.

S.No	FAM (TOXO)	Yakima Yellow/ VIC/HEX (IC)	<div style="display: flex; align-items: center;"> <span style="margin-right: 10px;">← Fluorophore</span> <div style="border-bottom: 1px solid black; width: 100%;"></div> </div> Interpretation	Conclusion
1	✓	✓	TOXO DNA detected	Proceed for further Analysis
2	✓	-		
3	-	✓	TOXO DNA not detected	
4	-	-	Possible inhibition of PCR	Dilute the DNA sample (1:10) and repeat the Assay

**Note: All the Target channels (FAM and Yakima Yellow/ VIC/HEX) to be analyzed individually.**

**Validated Instruments**

- Thermo QS5 Real-Time PCR System
- Bio-Rad™ CFX 96



HLSS Manufacturing Pvt Ltd  
 Plot No's M14, M15, M16, TSIIC Medical device park  
 Sultanpur village, Ameenpur Mandal,  
 SangareddyDist, TS-502319