

Medix Biochemica

Product Manual
Cat. No: #8701

Isotherm3G DNA Polymerase

Description

Isotherm3G DNA Polymerase is a mesophilic DNA polymerase, which synthesizes DNA from both DNA and RNA templates with a high strand displacement activity. It exhibits 5' to 3' polymerase activity but lacks any nuclease activity. Isotherm3G DNA Polymerase is the next generation of the original Isotherm DNA polymerase with further improved reverse transcription activity.

The product components have been optimized and are perfectly suited for loop-mediated isothermal amplification (LAMP) at constant temperature. The optimal reaction temperature is 65°C. However, the enzyme is also active at lower and higher temperatures (60–68°C). The enzyme can be inactivated at temperatures higher than 70°C. Addition of an intercalating dye allows the reaction to be monitored using a real-time PCR instrument. Isotherm3G DNA Polymerase is not suitable for use in PCR.

Isotherm3G DNA Polymerase utilizes an aptamer-based warm-start feature to prevent false amplification at low temperatures. For more information, please contact us.

Kit Components

Component	S pack*	M pack*
Isotherm3G DNA Polymerase (8 U/μL)	1 x 200 μL	1 x 1 mL
10x Isotherm3G reaction buffer	1 x 1 mL	3 x 1 mL

*Other pack sizes, bulk orders and customization are available upon request.

Storage and Shipment

Transport with ice pack. The reagents should be stored at -20°C upon arrival. The reagents are stable until the expiration date if stored correctly.

Reaction Master Mix Set-Up for LAMP Assay

The recommended reaction master mix set-up for a 25 μL LAMP reaction volume is shown in the table below. Reactions should be setup on ice. Mix and spin down all solutions carefully before use. After the preparation of the master mix, incubate at 65°C for 30–60 minutes.

To get optimal performance, a temperature gradient experiment (suggested is 60–68°C) and titration of Mg²⁺ concentration (suggested is 4–10 mM final) can be performed.

Reagent	Volume (μL)	Final concentration
Isotherm3G DNA Polymerase (8 U/μL)	1	0.32 U/μL
10x Isotherm3G reaction buffer	2.5	1x
*Primer mix	Variable	Variable
dNTPs (10 mM)	3.5	1.4 mM
Magnesium chloride (100 mM)	1.625	6.5 mM
DNA/cDNA/RNA template	Variable	Variable
Nuclease-free Water	Up to 25 μL final volume	
Total volume	25 μL	

*Primer mixes and concentrations depend strongly on the employed isothermal reaction method. Typical primer concentration for LAMP: 1.6 μM FIP/BIP primers, 0.2 μM F3/B3 primers, 0.4 μM LoopF/B primers.