Medix Biochemica

Product Manual
Cat. No: #8601

Fast *Bst* RT Mix

Description

Fast *Bst* RT Mix is a simple ready-to-use mix containing recombinant DNA polymerase expressed by Geobacillus stearothermophilus (formerly Bacillus stearothermophilus). The DNA polymerase displays high strand displacement activities, exhibits 5' to 3' polymerase activity, but lacks 5' to 3' exonuclease activity. Fast *Bst* RT Mix is suitable for isothermal nucleic acid amplification methods such as loop-mediated isothermal amplification (LAMP). Fast *Bst* RT Mix also contains an efficient thermostable reverse transcriptase with an RNase inhibitor (RTase Amp) to prevent degradation of RNA templates by RNases. This allows simultaneous reverse transcription and amplification of RNA target templates.

Fast *Bst* RT Mix is tolerant to inhibitors, enabling rapid and robust LAMP reactions at a constant temperature. The typical reaction temperature is 65°C. However, the enzyme is also active at lower and higher temperatures (55–70°C). The enzyme can be inactivated at temperatures higher than 80°C. Addition of an intercalating dye allows the reaction to be monitored using a real-time PCR instrument. Reactions can also be run using small and portable instruments with incubation and fluorescence measurement capabilities.

Kit Components

Component	S pack*	M pack*
2x Fast *Bst* Mix	1.25 mL	4 x 1.6 mL
20x Fluorescent dye	0.125 mL	0.625 mL
RTase Amp	0.2 mL	1 mL

*Other pack sizes or bulk orders are available upon request.

Storage and Shipment

Transport with an 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

The recommended master mix set-up for a 25 µL reaction volume is shown in the table below. After preparation of the master mix, incubate at 65°C for 30 minutes. The reaction time can be extended, and the incubation temperature can be varied between 55°C and 70°C to improve sensitivity and speed. The reaction can be monitored in a qPCR instrument by measuring fluorescence (FAM) every 10–30 seconds.

Reagent	Volume (µL)	Final concentration
2x Fast *Bst* Mix	12.5	1x
20x Fluorescent dye	1.25	1x
RTase Amp	2	1x
∞10x LAMP primer set	2.5	1x
RNA/DNA/cDNA template	X	Variable
Nuclease-free Water	Up to 25 µL final volume	

∞LAMP primers should be designed using an appropriate primer design tool. A predicted melting temperature of around 60˚C is recommended. The 10x primer set should contain 16 µM FIP, 16 µM BIP, 2 µM F3, 2 µM B3, 4–8 µM LoopF, and 4–8 µM LoopB in TE buffer or water.

Technical information and support

For technical enquiries or assay development support, please contact us via e-mail at: mdx@medixbiochemica.com.

Additional information and technical resources are available on our website at: info.medixbiochemica.com/resources.