Medix Biochemica

Product Manual
Cat. No: #8201

Fast *Bst* Polymerase Hot-Start

Description

Fast *Bst* Polymerase Hot-Start is a recombinant DNA polymerase expressed by Geobacillus stearothermophilus (formerly Bacillus stearothermophilus) combined with aptamer-based heat-activation technology. The aptamer reversibly inhibits Bst Polymerase at room temperature, minimizing non-specific amplification until temperatures reach 45-50°C.

The DNA polymerase displays high strand displacement activities, exhibits 5' to 3' polymerase activity, but lacks 5' to 3' exonuclease activity. Fast *Bst* Polymerase Hot-Start is suitable for several nucleic acid amplification methods such as loop-mediated isothermal amplification (LAMP), strand invasion-based amplification (SIBA), whole genome amplification, multiple displacement amplification, and isothermal amplification. The enzyme is glycerol-free.

Fast *Bst* Polymerase Hot-Start 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. This product is not suitable for PCR.

Kit Components

Component	S pack*	M pack*
Fast *Bst* Polymerase Hot-Start	0.2 mL	1 mL
∞ 10x Fast Buffer A	0.5 mL	2 x 1.25 mL
∞∞ 5x Fast Buffer B	1 mL	3 x 1.7 mL

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

∞The 10x Fast Buffer A has been formulated for robust performance. The buffer contains MgSO4, dNTPs, enhancers, and stabilizers.

∞∞The 5x Fast Buffer B contains an additional enhancer to further improve the reaction speed

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. It is recommended to aliquot the enzyme at the first use to avoid excess freeze/thaws.

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
10x Fast Buffer A	2.5	1x
5x Fast Buffer B	5	1x
20x Fluorescent dye (optional)	1.25	1x
Fast *Bst* Polymerase Hot-Start	1	8 U
ΔLAMP primer set	2.5	1x
DNA/cDNA template	X	Variable
Nuclease-free Water	Up to 25 µL final volume	

ΔLAMP 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.