

TargetEx Robust Bst Polymerase Lyo-Ready 5 mg/mL

Catalogue Number: TGX-028-BS/-BM/-BL

INTRODUCTION

TargetEx Robust Bst Polymerase is a proprietary genetically modified version of the large fragment of a thermostable enzyme derived from the bacterium *Geobacillus stearothermophilus*, designed for increased velocity, salt tolerance and remarkable stability at ambient conditions. This enzyme is ideal for highly efficient isothermal amplifications such as Loop-mediated Isothermal Amplification (LAMP).

PREPARATION

Prepare a 10-fold enzyme dilution from the 5 mg/mL polymerase stock with 10 mM Tris, 100 mM KCl, 0.1 mM EDTA, pH 7.5 buffer. Use 1 μ L of this enzyme dilution for a 25 μ L LAMP reaction.

TYPICAL REACTION ASSEMBLY

To test the enzyme in a LAMP reaction, prepare a 10X Primer Mix using the following components with the suggested final concentrations: FIP (16 μ M), BIP (16 μ M), F3 (2 μ M), B3 (2 μ M), LoopF (4 μ M), and LoopB (4 μ M). Mix all the listed components (see below), then remove all visible bubbles by centrifuging the plate. Create a heat profile on your thermal cycler according to the thermal cycler program setup below (in LAMP applications using PCR machines, fluorescence is often measured cyclically with 15 s incubation at 65 $^{\circ}$ C followed by readout, repeated up to 60 times). Select SYBR channel for fluorescence readout when using Syto9 stain. For reliable results, we recommend including technical replicates and obtaining a melt curve.

Store the product at -20 $^{\circ}$ C. Avoid freeze-thaw cycles.

COMPONENT LIST

Materials	Volume to 25 μ L (single measurement)	Recommended final concentration
Nuclease-Free Water (NFW)	11.5 μ L	N/A
10X Reaction Buffer	2.5 μ L	1X
10X Primer Mix	2.5 μ L	1X
dNTP mix (25 mM each)	1.5 μ L	1.5 mM
DNA Template	5 μ L	Variable
Fluorescent dye (e.g.: Syto9)	1 μ L	5 μ M
10X diluted TargetEx Robust Bst Polymerase Lyo-Ready 5 mg/mL	1 μ L	~8 U/reaction

THERMAL CYCLER PROGRAM SETUP

No.	Step	Temperature ($^{\circ}$ C)	Duration	Number of Cycles
1	Amplification	65	15 sec	60
2	Inactivation	95	2 min	1
3	Melt Curve	60-95	Variable	Variable

If you need further information or would like to try our products, please do not hesitate to contact us at info@targetex.com.

