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Bst 2.0 DNA Polymerase

Cat. No. : CW3323S (1600 U)
Storage Condition: -20°C

Components

Component	CW3323S 1600 U
BST 2.0 DNA Polymerase (8 U/μL)	200 μL
10×Bst 2.0 Reaction Buffer	1.5 mL
100mM $MgSO_4$ Solution	1.5 mL

Introduction

Bst 2.0 DNA Polymerase is a recombinase expressed and purified by *E. coli.* The gene is derived from *Bacillus stearothermophilus* with partial point mutations based on the original sequence. This protein has stronger 5'→3' DNA polymerase activity, strand displacement activity, reverse transcription activity, and no 5'→3' exonuclease activity. It is used in DNA loop-mediated isothermal amplification (LAMP), multiple displacement amplification (MDA), whole genome amplification (WGA), etc.

Activity Definition

The amount of enzyme required to incorporate 10 nmol deoxynucleotides into acidic insoluble substances at 65 °C for 30 minutes is defined as 1 active unit (U).

Heat Inactivation

This product can be inactivated after incubation at 80 °C for 5 min.

Quality Control

After multiple column purifications, SDS-PAGE test showed that the purity of the product was more than 98%, and no exogenous nuclease activity was detected.

Protocol

The following components are mixed proportionally and incubated at 65 °C for 30-60 min, and incubated at 80 °C for 5 min to inactivate.

Component	25 μL system	Final concentration
10×Bst 2.0 Reaction Buffer	2.5 μL	1× (with 2 mM $MgSO_4$)
100 mM $MgSO_4$ Solution	1.5 μL	6 mM (8 mM total)
dNTP Mix (10 mM)	3.5 μL	1.4 mM each
FIP/BIP Primers (25×)	1 μL	1.6 μM
F3/B3 First (25×)	1 μL	0.2 μM
LoopF/B Primers (25×)	1 μL	0.4 μM
Bst 2.0 DNA Polymerase (8 U/μL)	0.5-1 μL	160-320 U/mL
DNA Sample	Variable	>10 copies or more
Sterile water	To 25 μL	
Total	25 μL	

Note:1) LAMP primers consist of 4 or 6 (including Loop) primers, 25 × primers include: 40 μM FIP, 40 μM BIP, 5 μM F3, 5 μM B3, 10 μM LoopF, 10 μM LoopB.
2) If the reaction needs to be optimized, the Mg^{2+} concentration (4-10 mM), the amount of enzyme or the reaction temperature (60-72 °C) can be adjusted. The optimal reaction temperature of the enzyme is 65-68 °C.
3) Do not shake vigorously. Vigorous shaking may inactivate the enzyme.
4) After mixing, ensure that there are no bubbles in the reaction system.