Tinzyme Co., Limited

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BST 2.0 DNA Polymerase

Product Number: BS05

Shipping and Storage

-20℃.

Components

Component	BS05	BS05	BS05
BST 2.0 DNA Polymerase (8U/μl)	200μl	200μl×5	-
BST 2.0 DNA Polymerase (32U/μl)	-	-	1.25ml
10×BST 2.0 Buffer with Mg^{2+}*	1ml	1ml×5	10ml
100mM MgSO4	500μl	500μl×5	5ml

*The reaction buffer contains 40mM Mg

Description

BST 2.0 DNA Polymerase is a homolog of Bacillus stearothermophilus DNA polymerase (BST DNA Polymerase, Large Fragment), derived from E.coli strain. Obtained through multiple purification and isolation after expression in Escherichia coli using gene recombination technology. This enzyme has 5′→ 3′ DNA polymerase activity, but lacks 5′→ 3′ exonuclease activity. Compared with wild-type BST DNA polymerase large fragments, BST 2.0 DNA polymerase exhibits higher amplification speed, yield, and sensitivity.

Features

1. Compared with large fragments of BST DNA polymerase, the amplification speed and sensitivity have been significantly improved.
2. Optimization was carried out for loop mediated isothermal DNA amplification (LAMP).
3. Strong chain displacement activity.

Application

1. DNA sequencing rich in GC sequences;
2. Rapid sequencing of trace (nanogram) DNA templates;
3. Random primer DNA labeling;
4. Double stranded DNA 5 'protruding end patch labeling method;
5. It can be used for isothermal DNA amplification, such as loop mediated isothermal amplification (LAMP), whole genome amplification (WGA), helicase isothermal gene amplification (HDA), etc.

Unit definition

The amount of enzyme required to add 25nmol of dNTPs to acid insoluble precipitate within 30 minutes at 65 ℃ is defined as one active unit.

Protocol

LAMP commonly used reaction systems (20μL)

Pre mix the following reagents on ice and react at 60-65 ℃ for 30-60 minutes.

Component	Volume	Final Conc.
10×BST 2.0 Buffer with Mg^{2+}	2μl	1×
100mM $MgSO_4$	0.8μl	4mM[1)]

dNTPs (10mM each)	2.8μl	1.4mM
10×Primer[2)]	2μl	1×
BST 2.0 DNA Polymerase	6.4U	0.32U/μl
Template	Xμl	≥10fg
Nuclease-free Water	Up to 20μl	

Note:1)The final concentration of Mg^{2+} in the system is 8mM;

2)10×Primer contains 2μM F3/B3, 16μM FIP/BIP and 4μM LF/LB (adding ring primers can significantly shorten reaction time).

Note

1. Thermal deactivation condition: reaction at 80 ℃ for 10 minutes.
2. It is recommended to operate the three items of reagent premixing, template addition, and detection in different spaces as much as possible to avoid contamination of reagents and other substances that may affect subsequent experiments;
3. Suggest using dUTP/UDG pollution prevention system to prevent aerosol pollution;
4. The reaction is greatly affected by Mg^{2+} or enzymes. If the reaction result is not ideal, the final concentration of Mg^{2+} can be adjusted to 10mM for experimentation. If necessary, the final concentration of Mg^{2+} in the reaction system can be adjusted to 6-12 mM or the BST 2.0 DNA Polymerase activity can be adjusted to 0.16-0.64 U/μl activity to achieve optimal results;
5. 2-3 sets of primers can be designed simultaneously, and the best set can be selected for experimentation;
6. Fluorescent dyes such as Eva Green can be added to the reaction for real-time detection;
7. After the reaction is completed, if opening the lid, it is necessary to perform thermal deactivation before opening the lid for testing.