

Product components

Components	Component number	Size-1	Size-2
		1600 U	8000 U
Ruby Bst DNA Polymerase(8,000 U/mL)	RM20553	200 µL	1 mL
10X Isothermal Amplification Buffer II	RM20832	1.25 mL	1.25 mL*2
MgSO4 (100 mM)	RM20143	1.25 mL	1.25 mL*2

Product Description

Ruby Bst DNA Polymerase is an in silico designed homologue of *Geobacillus stearothermophilus* DNA Polymerase I (large fragment). Ruby Bst DNA Polymerase has 5'→3' DNA polymerase activity and strong strand displacement activity but lacks 5'→3' exonuclease activity. Ruby Bst DNA Polymerase displays improved amplification speed, yield, salt tolerance and thermo stability compared with wild-type Bst DNA Polymerase, Large Fragment.

Source

Ruby Bst DNA Polymerase is prepared from an *E. coli* strain.

Applications

Isothermal Amplification: Loop-Mediated Isothermal Amplification (LAMP)

Definition of Activity Unit

One unit of the enzyme catalyzes the incorporation of 25 nmol of dNTP into acid-insoluble precipitate in 30 minutes at 65°C.

Store

-20°C, avoid repeated freezing and thawing.

Inhibition and Inactivation

Inactivate at 80 °C for 20 min.

Other Materials

Reagent: dNTP Mix, FIP/BIP Primers, F3/B3 Primers, LoopF/LoopB Primers, Nuclease-free Water.

Instrument: PCR instrument or water bath pot.

Protocol

1. LAMP

Reagent	Volume	Final concentration
10X Isothermal Amplification Buffer II	2.5 µl	1X (contains 2 mM MgSO4)
MgSO4 (100 mM)	1.5 µl	6 mM
dNTP Mix (10 mM)	3.5 µl	1.4 mM each
Primers (25X)	1 µl	1.6 µM FIP/BIP, 0.2 µM F3/B3, 0.4 µM Loop F/B
Ruby Bst DNA Polymerase(8,000 U/mL)	1 µl	/
DNA Sample	variable	/
Nuclease-free Water	to 25 µl	

2. Mix the reaction components and centrifuge briefly to collect the contents at the bottom of the tube.

3. Incubate the mixture at 65 °C for 30-60 min.
4. Stop the reaction by heating at 80 °C for 20 min.

Precautions

- 1) It is recommended to operate reagent premix, template addition, and detection in different spaces to avoid reagent contamination, so that it would affect subsequent experiments;
- 2) The reaction is greatly affected by Mg^{2+} or enzyme. If the reaction result is not ideal, the final concentration of Mg^{2+} can be adjusted between 4mM and 10 mM. If necessary, Bst enzyme activity can be titrated to achieve ideal performance;
- 3) It is recommended to design 2-3 pairs of primers at the same time, and select the best one for experiments;
- 4) Use NTC(no template control) to detect the specificity of amplification;
- 5) If it is necessary to open the lid after the reaction is completed, it is necessary to perform thermal deactivation before opening the lid.
- 6) For your safety and health, please wear lab coat and disposable gloves.