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Limited liability company

Bst DNA-polymerase

Cat. Number: E-10002, E-10010

Product description

DNA-polymerase LF Bst is a large Bst polymerase fragment of *Bacillus stearothermophilus*. Enzyme contains histidine mark on its C-terminus and has molecular mass of 68,9 kDa. The enzyme is high-processive and catalyses 5'-> 3' DNA synthesis. Fragment lacks 5'-> 3' and 3'-> 5'-exonuclease activity as well as 5'-3' displacement activity.

Optimal activity conditions for the enzyme are 65 °C and pH 8,9.

Application:

- loop isothermal amplification (LAMP)
- Whole-genome sequencing

Source:

Bst DNA-polymerase was obtained from *E.coli* strain, carrying plasmid with cloned full-size gene of *Bacillus stearothermophilus* DNA-polymerase I large fragment.

Activity units:

One activity unit corresponds to the enzyme amount required for inclusion of 10 nmoles dNTPs in non-acid-soluble DNA fraction in 0 min at 65°C.

Quality control:

Every enzyme batch is tested for endonuclease and non-specific exonuclease activity absence, enzyme sensitivity.

Enzyme concentration: 10 UA/ μ l.

Storage buffer: to the amout of enzyme 20 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.1%

Triton X-100, 50% glycerin.

10x LAMP guffer (Cat №: SP030, supplied separately):

300 mM Tris-HCl (pH 8.9), 50 mM (NH₄)₂SO₄, 0.5 mg/ml BSA, 2.0% Tween 20.

Typical LAMP assay:

Reaction mixture contains:

Component	Final concentration	Volume, μl
10× LAMP buffer (Cat №: SP030)	1×	5
100 mM MgSO4	4-10 mM (6 mM)*	3*
10 mM dNTP Mix (Kat №: NM10)	1.4 mM each	7
16 μM FIP/BIP Primers	1.6 μΜ	5
2 μM F3/B3 Primers	0.2 μΜ	5
8 μM LoopF/B Primers	0.8 μΜ	5
10 U/μl <i>Bst</i> DNA-polymerase	0.008 – 0.2 10 U/µl (0.033 U/µl)*	0.2*
DNA-matrix	> 10 copies per reaction	variable
Sterile water (Cat. №: SP010)		up to 50
Total reaction volume	50 μΙ	

^{*} recommended concentration

To perform amplification specialaised isothermal reaction amplifiers as well as PCR amplifiers. It is recommended to perform amplification reaction at the 60 - 65 $^{\circ}\text{C}$ temperature range with the duration of 20–30 minutes. For the correct results interpretation it is recommended to always set a control reaction without the DNA-matrix.

Transportation: in thermostated containers with cooling elements, tolerating temperature increment up to environment temperature, if transported in 10 days.

Storage conditions and period: 1 year at -20 °C.