Datasheet

Bst DNA Polymerase, Exonuclease Minus

Features:
- reverse transcription activity
- increased activity

Applications:
- nucleic acid amplification methods, including isothermal amplification
- whole genome amplification
- multiple displacement amplification
- sequencing DNA with high GC content and secondary structures
- rapid sequencing from nanogram amounts of DNA Template

Description:
Bst DNA Polymerase, Exonuclease Minus, is a 67 kDa *Bacillus stearothermophilus* DNA Polymerase protein (large fragment) which has a 5'-3' polymerase activity and strand displacement activity but lacks 3' – 5' exonuclease activity. Also has reverse transcription activity.

Unit definition:
One unit catalyzes the incorporation of 10 nmol of dNTP into acid-insoluble material in 30 minutes at 65°C in 20 mM Tris-HCl pH 8.8, 10 mM $(NH_4)_2SO_4$,10 mM KCl, 2 mM $MgSO_4$, 0.1% Triton X-100, 30 nM M13mp18 ssDNA, 70 nM M13 sequencing primer(-47) 24 mer 200 µM dGTP, dATP, dTTP, dCTP (a mix of unlabeled and [^{33}P]dCTP) and 0.1 mg/ml BSA.

Heat inactivation 80°C for 20 minutes.

Concentration: up to 8 U / µl

Reaction buffer (10X): 200 mM Tris-HC1(pH8.8), 100 mM $(NH_4)_2SO_4$, 100 mM KCl, 20 mM $MgSO_4$, and 1.0 % Triton X-100

Qualtity Tests:
Endonuclease Activity: Incubation of 8 units and 50 units of enzyme with 1µg of supercoiled pBR322 DNA for 16 hours at 37° and 65°C resulted in no detectable conversion to relaxed or linear forms by agarose gel electrophoresis.

Exonuclease Activity: Incubation of 8 units and 50 units of enzyme with 1µg of HIND III-cut lambda DNA for 16 hours at 37° and 65°C resulted in no smearing of bands on agarose gels. Single stranded and double stranded exonuclease activites were tested by incubating 10 µl of enzyme with radiolabeled DNA substrate for one hour at 37° and 65°C, resulting in less than 0.1% release of TCA-soluble counts.

Purity: >99% by SDS PAGE. No detectable DNA contamination. 10 µl of the enzyme was tested for *E.coli* genomic DNA contamination by PCR amplifying with the *E.coli* 16S ribosomal primers.

Note:
- Recommended for long term storage: 0.1% Triton X-100
- Reaction temperatures above 70°C are not recommended.
- Bst DNA Polymerase cannot be used for thermal cycle sequencing.

.. a good decision ..

References:

1. Stenesh, J. und Roe, B.A. (1972) Biochim. Biophys. Acta. 272, 156-166.
2. Hugh, G. und Griffin, M. (1994) PCR Technology, p.p.228-229.
3. McClary, J. et al. (1991) J. DNA Sequencing and Mapping, p.p.173-180.
4. Tomita N. et al. (2008) Nature Protocols, p.p. 877-882

Storage: -20°C
Transport: on blue ice

Ordering information:

Cat.-no	Description	Amount
A600	Bst DNA Polymerase	2000 units
A610	Bst DNA Polymerase	10000 units