Bst 2.0 WarmStart™ DNA Polymerase

1-800-632-7799
info@neb.com
www.neb.com

M0538S 003131215121

M0538S

1,600 units **8,000 U/ml** **Lot: 0031312**

RECOMBINANT **Store at –20°C** **Exp: 12/15**

Description: *Bst* 2.0 WarmStart DNA Polymerase is an *in silico* designed homologue of *Bacillus stearothermophilus* DNA Polymerase I, Large Fragment (*Bst* DNA Polymerase, Large Fragment) with a reversibly-bound aptamer, which inhibits polymerase activity at temperatures below 45°C. The aptamer rapidly releases the *Bst* 2.0 WarmStart DNA Polymerase above 45°C and therefore no special activation step is needed to activate the polymerase. *Bst* 2.0 WarmStart DNA Polymerase contains 5´→3´ DNA polymerase activity and strong strand-displacement activity but lacks 5´→3´ exonuclease activity. *Bst* 2.0 WarmStart DNA Polymerase displays improved amplification speed, yield, salt tolerance, and thermostability compared to wild-type *Bst* DNA Polymerase, Large Fragment.

Source: *Bst* 2.0 WarmStart DNA Polymerase is prepared from an *E. coli* strain that expresses the *Bst* 2.0 DNA Polymerase protein from an inducible promoter.

Applications:

- Isothermal DNA amplification
- Applications requiring strand-displacement DNA synthesis
- DNA sequencing through high GC regions
- Rapid sequencing from nanogram amounts of DNA template

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM dithiothreitol, 0.1% Triton® X-100 and 50% glycerol.

Reagents Supplied with Enzyme:
Isothermal Amplification Buffer (10X)

Reaction Conditions:
Specific reaction conditions will vary for different isothermal amplification applications. For best results, use 1X Isothermal Amplification Buffer.
Incubate at 65°C.

1X Isothermal Amplification Buffer:
20 mM Tris-HCl
10 mM $(NH_4)_2SO_4$
50 mM KCl
2 mM $MgSO_4$
0.1% Tween® 20
pH 8.8 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme that will incorporate 25 nmol of dNTP into acid insoluble material in 30 minutes at 65°C.

Unit Assay Conditions: 50 mM KCl, 20 mM Tris-HCl (pH 8.8), 10 mM $MgCl_2$, 30 nM M13mp18 SS DNA, 70 nM M13 sequencing primer (–47) 24 mer, 200 µM dATP, 200 µM dCTP, 200 µM dGTP, 100 µM dTTP including [^{3}H]-dTTP and 100 µg/ml BSA.

Heat Inactivation: 80°C for 20 minutes.

Quality Control Assays

Exonuclease Assay: Incubation of a 50 µl reaction in 1X ThermoPol® Reaction Buffer containing a minimum of 500 units of *Bst* 2.0 DNA Polymerase with 1 µg of a mixture of single and double-stranded [^{3}H] *E. coli* DNA (10^5 cpm/µg) for 4 hours at 65°C releases < 0.1% of the total radioactivity.

Endonuclease Assay: Incubation of a 50 µl reaction in 1X ThermoPol Reaction Buffer containing a minimum of 500 units of *Bst* 2.0 DNA Polymerase with 1 µg of supercoiled ϕX174 DNA for 4 hours at 65°C results in < 10% conversion to the nicked form as determined by agarose gel electrophoresis.

Physical Purity: Purified to > 99% homogeneity as determined by SDS-PAGE analysis using Coomassie Blue detection.

(see other side)

CERTIFICATE OF ANALYSIS

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(see other side)

CERTIFICATE OF ANALYSIS

Phosphatase Assay: Incubation of a 200 µl reaction in 1 M Diethanolamine (pH 9.8) and 0.5 mM $MgCl_2$ containing 2.5 mM *p*-Nitrophenol Phosphate and a minimum of 100 units of *Bst* 2.0 DNA Polymerase incubated for 4 hours at 37°C yields no detectable phosphatase activity as determined by spectrophotometric analysis of released *p*-nitrophenylene anion at 405 nm.

RNase Activity: Incubation of a 10 µl reaction in 1X NEBuffer 4 containing a minimum of 1 µl of *Bst* 2.0 WarmStart DNA Polymerase and 40 ng of F-300 RNA transcript incubated for 16 hours at 37°C results in < 10% substrate degradation as determined by gel electrophoresis using fluorescent detection.

Enzyme Properties

Activity in NEBuffers

ThermoPol Buffer	125%
Unit Assay Conditions	100%
NEBuffer 1	25%
NEBuffer 2	100%
NEBuffer 3	100%
NEBuffer 4	100%
NEBuffer EcoRI	100%

Notes on Use: *Bst* 2.0 WarmStart DNA Polymerase does not exhibit 3´→ 5´ exonuclease activity.

Reaction temperatures above 70°C are not recommended.

Bst 2.0 WarmStart DNA Polymerase cannot be used for thermal cycle sequencing or PCR.

Companion Products Sold Separately:

Bst 2.0 DNA Polymerase

#M0537S	1,600 units
#M0537L	8,000 units
#M0537M	8,000 units

Bst DNA Polymerase, Large Fragment

#M0275S	1,600 units
#M0275L	8,000 units
#M0275M	8,000 units

Magnesium Sulfate ($MgSO_4$) Solution

#B1003S	6.0 ml

Isothermal Amplification Buffer Pack

#B0537S	6.0 ml

Deoxynucleotide Solution Set

#N0446S	25 µmol each

Deoxynucleotide Solution Mix

#N0447S	8 µmol each
#N0447L	40 µmol each

Phosphatase Assay: Incubation of a 200 µl reaction in 1 M Diethanolamine (pH 9.8) and 0.5 mM $MgCl_2$ containing 2.5 mM *p*-Nitrophenol Phosphate and a minimum of 100 units of *Bst* 2.0 DNA Polymerase incubated for 4 hours at 37°C yields no detectable phosphatase activity as determined by spectrophotometric analysis of released *p*-nitrophenylene anion at 405 nm.

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#M0537M	8,000 units

Bst DNA Polymerase, Large Fragment

#M0275S	1,600 units
#M0275L	8,000 units
#M0275M	8,000 units

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Isothermal Amplification Buffer Pack

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Deoxynucleotide Solution Set

#N0446S	25 µmol each

Deoxynucleotide Solution Mix

#N0447S	8 µmol each
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TWEEN® is a registered trademark of Uniqema Americas LLC.

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