

Bst 2.0 WarmStart™ DNA Polymerase



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



M0538S 003140616061

M0538S



1,600 units **8,000 U/ml** **Lot: 0031406**

RECOMBINANT **Store at -20°C** **Exp: 6/16**

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)

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Reagents Supplied with Enzyme:

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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 (NH₄)₂SO₄
50 mM KCl
2 mM MgSO₄
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 MgCl₂, 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 [³H]-dTTP and 100 μg/ml BSA.

Heat Inactivation: 80°C for 20 minutes.

(see other side)

CERTIFICATE OF ANALYSIS

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CERTIFICATE OF ANALYSIS

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 [³H] *E. coli* DNA (10⁵ 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.

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

Phosphatase Assay: Incubation of a 200 µl reaction in 1 M Diethanolamine (pH 9.8) and 0.5 mM MgCl₂ 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:

<i>Bst</i> 2.0 DNA Polymerase	
#M0537S	1,600 units
#M0537L	8,000 units
#M0537M	8,000 units
<i>Bst</i> 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



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#M0537L	8,000 units
#M0537M	8,000 units

<i>Bst</i> 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



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