Bst 2.0 WarmStart™ DNA Polymerase

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

**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 (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 [³²P]-dTTP and 100 µg/ml BSA.

**Heat Inactivation:**
80°C for 20 minutes.

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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

<table>
<thead>
<tr>
<th>Activity in NEBuffers</th>
<th>Activity in NEBuffers</th>
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<th>Activity in NEBuffers</th>
<th>Activity in NEBuffers</th>
</tr>
</thead>
<tbody>
<tr>
<td>ThermoPol Buffer</td>
<td>125%</td>
<td>Unit Assay Conditions</td>
<td>100%</td>
<td>NEBuffer 1</td>
</tr>
<tr>
<td>NEBuffer 1</td>
<td>25%</td>
<td>NEBuffer 2</td>
<td>100%</td>
<td>NEBuffer 3</td>
</tr>
<tr>
<td>NEBuffer 2</td>
<td>100%</td>
<td>NEBuffer 4</td>
<td>100%</td>
<td>NEBuffer EcoRI</td>
</tr>
</tbody>
</table>

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:

<table>
<thead>
<tr>
<th>Bst 2.0 DNA Polymerase</th>
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</tr>
</thead>
<tbody>
<tr>
<td>#M0537S</td>
<td>1,600 units</td>
<td>#M0537L</td>
</tr>
<tr>
<td>#M0537L</td>
<td>8,000 units</td>
<td>#M0537S</td>
</tr>
<tr>
<td>#M0537M</td>
<td>8,000 units</td>
<td>#M0275S</td>
</tr>
<tr>
<td>#M0275S</td>
<td>1,600 units</td>
<td>#M0275L</td>
</tr>
<tr>
<td>#M0275M</td>
<td>8,000 units</td>
<td>#M0275F</td>
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<tr>
<td>Magnesium Sulfate (MgSO₄) Solution</td>
<td>#B1003S 6.0 ml</td>
<td>#B0537S</td>
</tr>
<tr>
<td>Isothermal Amplification Buffer Pack</td>
<td>#N0446S 25 µmol each</td>
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</tr>
<tr>
<td>Deoxynucleotide Solution Mix</td>
<td>#N0447L 40 µmol each</td>
<td></td>
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</table>