

## New England Biolabs Product Specification

<i>Product Name:</i>	<i>Deep Vent® (exo-) DNA Polymerase</i>
<i>Catalog #:</i>	<i>M0259S/L/V</i>
<i>Concentration:</i>	<i>2,000 units/ml</i>
<i>Unit Definition:</i>	<i>One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid-insoluble material 30 minutes at 75°C.</i>
<i>Shelf Life:</i>	<i>24 months</i>
<i>Storage Temp:</i>	<i>-20°C</i>
<i>Storage Conditions:</i>	<i>10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.1 % Triton®X-100, 50 % Glycerol, (pH 7.4 @ 25°C)</i>
<i>Specification Version:</i>	<i>PS-M0259S/L v2.0</i>
<i>Effective Date:</i>	<i>12 Feb 2020</i>

### Assay Name/Specification (minimum release criteria)

**Endonuclease Activity (Nicking)** - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of Deep Vent™ (exo-) DNA Polymerase incubated for 4 hours at 37°C and 75°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

**Exonuclease Activity (Radioactivity Release)** - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] *E. coli* DNA and a minimum of 20 units of Deep Vent™ (exo-) DNA Polymerase incubated for 4 hours at 37°C and 75°C releases <0.1% of the total radioactivity.

**Non-Specific DNase Activity (16 Hour)** - A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 2 units of Deep Vent™ (exo-) DNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

**PCR Amplification (2.0 kb Lambda DNA)** - A 25 µl reaction in ThermoPol® Reaction Buffer in the presence of 200 µM dNTPs and 0.2 µM primers containing 5 ng Lambda DNA with 1 unit of Deep Vent™ (exo-) DNA Polymerase for 25 cycles of PCR amplification results in the expected 2.0 kb product.

**Phosphatase Activity (pNPP)** - A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl<sub>2</sub> containing 2.5 mM *p*-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units Deep Vent™ (exo-) DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.



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<p><b>Protein Purity Assay (SDS-PAGE)</b> - Deep Vent™ (exo-) DNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>
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<p><b>Single Stranded DNase Activity (FAM-Labeled Oligo)</b> - A 20 µl reaction in ThermoPol® Reaction Buffer containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 20 units of Deep Vent™ (exo-) DNA Polymerase incubated for 30 minutes at 37°C and 75°C yields &lt;10% degradation as determined by capillary electrophoresis.</p>
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Date 12 Feb 2020

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Derek Robinson  
Director, Quality Control

