New England Biolabs
Product Specification

Product Name: Deep Vent® DNA Polymerase
Catalog #: M0258S/L
Concentration: 2,000 units/ml
Unit Definition: One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid insoluble material in 30 minutes at 75°C.
Shelf Life: 24 months
Storage Temp: -20°C
Storage Conditions: 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)
Specification Version: PS-M0258S/L v2.0
Effective Date: 23 Jan 2018

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking, Polymerase, dNTP) - A 50 µl reaction in ThermoPol® Reaction Buffer in the presence of 400 µM dNTPs containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of Deep Vent® DNA Polymerase incubated for 4 hours at 37°C results in <10% conversion to the nicked form 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 0.5 units of Deep Vent® DNA Polymerase for 30 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 MgCl₂ containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of Deep Vent® DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.

Protein Purity Assay (SDS-PAGE) - Deep Vent® DNA Polymerase is ≥ 98% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

qPCR DNA Contamination (E. coli Genomic) - A minimum of 2 units of Deep Vent® DNA Polymerase is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.
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<table>
<thead>
<tr>
<th>Assay Name/Specification (minimum release criteria)</th>
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<tr>
<td><strong>RNase Activity (Extended Digestion)</strong> - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Deep Vent® DNA Polymerase is incubated at 37°C. After incubation for 16 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</td>
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**Derek Robinson**

Director of Quality Control

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Date 23 Jan 2018