New England Biolabs
Product Specification

Product Name: Deoxynucleotide (dNTP) Solution Set
Catalog #: N0446S/V
Concentration: 100 mM
Unit Definition: N/A
Shelf Life: 24 months
Storage Temp: -20°C
Storage Conditions: Supplied in Ultrapure water as a sodium salt (pH 7.5)
Effective Date: 16 Jan 2022

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in NEBuffer 2 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 1 µl of dATP, dCTP, dGTP, and dTTP incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

PCR Amplification (0.5 kb Lambda, dNTPs) - A 50 µl reaction in ThermoPol® Reaction Buffer in the presence of 200 µM dATP, dCTP, dGTP, and dTTP and 0.2 µM primers containing 1 ng Lambda DNA with 1.25 units of Taq DNA Polymerase for 25 cycles of PCR amplification results in the expected 0.5 kb product.

PCR Amplification (2.0 kb Lambda, dNTPs) - A 50 µl reaction in ThermoPol® Reaction Buffer in the presence of 200 µM dATP, dCTP, dGTP, and dTTP and 0.2 µM primers containing 1 ng Lambda DNA with 1.25 units of Taq DNA Polymerase for 25 cycles of PCR amplification results in the expected 2.0 kb product.

PCR Amplification (5.0 kb Lambda, dNTPs) - A 50 µl reaction in ThermoPol® Reaction Buffer in the presence of 200 µM dATP, dCTP, dGTP, and dTTP and 0.2 µM primers containing 1 ng Lambda DNA with 1.25 units of Taq DNA Polymerase for 25 cycles of PCR amplification results in the expected 5.0 kb product.

Physical Purity (HPLC) - dATP, dCTP, dGTP, and dTTP is ≥ 99% pure as determined by HPLC analysis.

RNase Activity (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl dATP, dCTP, dGTP, and dTTP is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.
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<table>
<thead>
<tr>
<th>Assay Name/Specification (minimum release criteria)</th>
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<tbody>
<tr>
<td><strong>Non-Specific DNase Activity (16 Hour)</strong> - 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 4 µl of dATP, dCTP, dGTP, and dTTP incubated for 16 hours at 37ºC results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</td>
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<tr>
<td><strong>Phosphatase Activity (pNPP)</strong> - 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 16 µl of dATP, dCTP, dGTP, and dTTP incubated for 4 hours at 37°C yields &lt;0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.</td>
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<td><strong>qPCR DNA Contamination (E. coli Genomic)</strong> - A minimum of 1 µl of dATP, dCTP, dGTP, and dTTP 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.</td>
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