

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

Product Name:	Therminator™ DNA Polymerase
Catalog #:	M0261S/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 $75^{\circ}$ C.
Lot #:	0201709
Assay Date:	09/2017
Expiration Date:	9/2019
Storage Temp:	-20°C
Storage Conditions:	10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version:	PS-M0261S/L v1.0
Effective Date:	11 Sep 2015

Assay Name/Specification (minimum release criteria)	Lot #0201709
<b>Endonuclease Activity (Nicking)</b> - A 50 $\mu$ l reaction in ThermoPol® Reaction Buffer containing 1 $\mu$ g of supercoiled PhiX174 DNA and a minimum of 20 units of Therminator <sup>TM</sup> DNA Polymerase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 $\mu$ l reaction in ThermoPol® Reaction Buffer containing 1 $\mu$ g of a mixture of single and double-stranded [ <sup>3</sup> H] <i>E. coli</i> DNA and a minimum of 20 units of Therminator <sup>TM</sup> DNA Polymerase incubated for 4 hours at either 37°C or 75°C releases <0.1% of the total radioactivity.	Pass
<b>Non-Specific DNase Activity (16 Hour)</b> - A 50 $\mu$ l reaction in NEBuffer 2 containing 1 $\mu$ g of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 2 units of Therminator <sup>TM</sup> 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.	Pass
<b>Phosphatase Activity (pNPP)</b> - A 200 $\mu$ l reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl <sub>2</sub> containing 2.5 mM <i>p</i> -Nitrophenol Phosphate (pNPP) and a minimum of 100 units Therminator <sup>TM</sup> DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> - Therminator <sup>TM</sup> DNA Polymerase is $\geq$ 98% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass



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Assay Name/Specification (minimum release criteria)	Lot #0201709
<b>RNase Activity (Extended Digestion)</b> - A 10 $\mu$ l reaction in NEBuffer 4 containing 40 ng of a 300 base single- stranded RNA and a minimum of 1 $\mu$ l of Therminator <sup>TM</sup> DNA Polymerase 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.	Pass

Authorized by Melanie Fortier 11 Sep 2015



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Inspected by Tony Spear-Alfonso 11 Sep 2017