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: DNA Polymerase I (E. coli)

Catalog #: M0209S/L
Concentration: 10,000 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 37°C.

 Lot #:
 0921608

 Assay Date:
 08/2016

 Expiration Date:
 8/2018

 Storage Temp:
 -20°C

Storage Conditions: 25 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-M0209S/L v1.0

Effective Date: 23 Sep 2015

Assay Name/Specification (minimum release criteria)	Lot #0921608
<b>Endonuclease Activity (Nicking)</b> - A 50 μl reaction in NEBuffer 2 containing 1 μg of supercoiled PhiX174 DNA and a minimum of 100 units of DNA Polymerase I ( <i>E. coli</i> ) incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Phosphatase Activity (pNPP)</b> - A 200 μl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl <sub>2</sub> containing 2.5 mM <i>p</i> -Nitrophenyl Phosphate (pNPP) and a minimum of 100 units DNA Polymerase I ( <i>E. coli</i> ) 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> - DNA Polymerase I ( <i>E. coli</i> ) is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
<b>qPCR DNA Contamination</b> ( <i>E. coli</i> <b>Genomic</b> ) - A minimum of 10 units of DNA Polymerase I ( <i>E. coli</i> ) is screened for the presence of <i>E. coli</i> genomic DNA using SYBR® Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is $\leq 1$ <i>E. coli</i> genome.	Pass
RNase Activity (Extended Digestion) - 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 DNA Polymerase I ( <i>E. coli</i> ) 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 23 Sep 2015







Inspected by Cathy Rezac 17 Aug 2016