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## New England Biolabs Certificate of Analysis

Product Name: DNA Polymerase I, Large (Klenow) Fragment

 Catalog #:
 M0210S/L

 Concentration:
 5,000 units/ml

 Lot #:
 0891603

 Assay Date:
 03/2016

 Expiration Date:
 3/2018

 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-M0210S/L v1.0

Effective Date: 16 Oct 2015

Assay Name/Specification (minimum release criteria)	Lot #0891603
Endonuclease Activity (Nicking) - A 50 $\mu$ l reaction in NEBuffer 2 containing 1 $\mu$ g of supercoiled PhiX174 DNA and a minimum of 50 units of DNA Polymerase I, Large (Klenow) Fragment 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, Large (Klenow) Fragment 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, Large (Klenow) Fragment 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 50 units of DNA Polymerase I, Large (Klenow) Fragment 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, Large (Klenow) Fragment 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 16 Oct 2015







Inspected by Karen Moreira 19 Feb 2016