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New England Biolabs Certificate of Analysis

Product Name: Klenow Fragment (3'-5' exo-)

Catalog Number: M0212L Concentration: 5,000 U/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.

Packaging Lot Number: 10058065 Expiration Date: 10/2021 Storage Temperature: -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-M0212S/L v1.0

Klenow Fragment (3'-5' exo-) Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
M0212LVIAL	Klenow Fragment (3'-5' exo-)	10055677	Pass	
B7002SVIAL	NEBuffer™ 2	10052180	Pass	

Assay Name/Specification	Lot # 10058065
Endonuclease Activity (Nicking) A 50 μl reaction in NEBuffer 2 containing 1 μg of supercoiled PhiX174 DNA and a minimum of 50 units of Klenow Fragment (3'–δ' exo-) incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Exonuclease Activity (Radioactivity Release) A 50 μl reaction in NEBuffer 2 containing 1 μg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 200 units of Klenow Fragment (3'—5' exo-) incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Non-Specific DNase Activity (16 Hour) A 50 μl reaction in NEBuffer 2 containing 1 μg of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 50 units of Klenow Fragment (3'–5' exo-) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Phosphatase Activity (pNPP)	Pass



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Assay Name/Specification	Lot # 10058065
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 Klenow Fragment (3'—5' exo-) 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) Klenow Fragment (3'—5' exo-) is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 50 units of Klenow Fragment (3'—5' exo-) 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.	Pass
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 of Klenow Fragment (3'—5' exo-) is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescence detection.	Pass
Single Stranded DNase Activity (FAM-Labeled Oligo) A 50 µl reaction in NEBuffer 2 containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 50 units of Klenow Fragment (3'—5' exo-) incubated for 30 minutes at 37°C yields <10% degradation as determined by fluorescent detection.	Pass

This product has been tested and shown to be in compliance with all specifications.

Production Scientist 12 Sep 2019 Minichiello

Packaging Quality Control Inspector

25 Nov 2019



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