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## New England Biolabs Certificate of Analysis

Product Name: Thermolabile Exonuclease I

Catalog Number: M0568L Concentration: 20,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme that will catalyze the

release of 2 nmol of acid-soluble nucleotide in a total reaction volume of 100  $\mu$ l in 6 minutes at 37°C in NEBuffer 3.1 with 0.17

mg/ml single-stranded [ ³H]-E.coli DNA.

Packaging Lot Number: 10094770
Expiration Date: 02/2023
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 250 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA,

50% Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-M0568S/L v1.0

Thermolabile Exonuclease I Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
M0568LVIAL	Thermolabile Exonuclease I	10094769	Pass	
B7203SVIAL	NEBuffer™ 3.1	10092686	Pass	

Assay Name/Specification	Lot # 10094770
RNase Activity Assay (4 Hour 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 Thermolabile Exonuclease I is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass
<b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 20 units of Thermolabile Exonuclease I 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
Protein Purity Assay (SDS-PAGE) Thermolabile Exonuclease I is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass



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Assay Name/Specification	Lot # 10094770
Non-Specific DNase Activity (16 Hour) A 50 μl reaction in CutSmart® Buffer containing 1 μg of PhiX174-HaeIII DNA and a minimum of 100 units of Thermolabile Exonuclease I incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Functional Testing (Thermolability) A 20 µl reaction in Standard Taq Reaction Buffer containing 20 pmol of 20-mer ssDNA and 20 units of Thermolabile Exonuclease I was incubated for 4 minutes at 37°C followed by heat inactivation for 1 minute at 80°C. The addition of 20 pmol of 20-mer ssDNA and incubation for 40 minutes at 37°C results in no cleavage of additional substrate as determined by capillary electrophoresis.	Pass
Endonuclease Activity (Nicking) A 50 μl reaction in CutSmart® Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 100 units of Thermolabile Exonuclease I incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Endonuclease Activity (Circular Single Stranded DNA) A 50 µl reaction in CutSmart® Buffer containing 1 µg of M13 single-stranded DNA and a minimum of 100 units of Thermolabile Exonuclease I incubated for 4 hours at 37°C results in <10% conversion to linear DNA as determined by agarose gel electrophoresis.	Pass

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

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John Greci Production Scientist 04 Feb 2021 Michael Tonello

Packaging Quality Control Inspector

04 Feb 2021



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