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: Antarctic Phosphatase

Catalog #: M0289S/L
Concentration: 5,000 units/ml

Unit Definition: One unit is defined as the amount of enzyme that will dephosphorylate 1 µg of pUC19 vector DNA cut with a restriction

enzyme generating 5´ recessed ends in 30 minutes at 37°C. Dephosphorylation is defined as >95% inhibition of

recircularization in a self-ligation reaction and is measured by transformation into E. coli.

 Lot #:
 0221603

 Assay Date:
 03/2016

 Expiration Date:
 3/2018

 Storage Temp:
 -20°C

Storage Conditions: 10 mM Tris-HCl, 1 mM MgCl₂, 0.01 mM ZnCl₂, 50 % Glycerol, (pH 7.4 @, 25°C)

Specification Version: PS-M0289S/L v1.0

Effective Date: 22 Jun 2016

Assay Name/Specification (minimum release criteria)	Lot #0221603
Endonuclease Activity (Nicking) - A 50 μl reaction in Antarctic Phosphatase Reaction Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 50 units of Antarctic Phosphatase 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 Antarctic Phosphatase Reaction Buffer containing 1 µg of a mixture of single and double-stranded [³ H] <i>E. coli</i> DNA and a minimum of 50 units of Antarctic Phosphatase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Non-Specific DNase Activity (16 Hour) - A 50 μ l reaction in NEBuffer 4 containing 1 μ g of PhiX174-HaeIII DNA and a minimum of 50 units of Antarctic Phosphatase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE) - Antarctic Phosphatase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (<i>E. coli</i> Genomic) - A minimum of 5 units of Antarctic Phosphatase 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 ≤ 1 <i>E. coli</i> genome.	Pass







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Assay Name/Specification (minimum release criteria)	Lot #0221603
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 Antarctic Phosphatase 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 Derek Robinson 22 Jun 2016







Inspected by Tanya Barshevsky 18 Mar 2016