

## New England Biolabs Certificate of Analysis

**Product Name:** Antarctic Phosphatase  
**Catalog Number:** M0289S  
**Concentration:** 5,000 U/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*.  
**Packaging Lot Number:** 10251935  
**Expiration Date:** 04/2026  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl, 1 mM MgCl<sub>2</sub>, 0.01 mM ZnCl<sub>2</sub>, 50 % Glycerol, (pH 7.4 @ 25°C)  
**Specification Version:** PS-M0289S/L v2.0

Antarctic Phosphatase Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0289SVIAL	Antarctic Phosphatase	10236900	Pass
B0289SVIAL	Antarctic Phosphatase Reaction Buffer	10208060	Pass

Assay Name/Specification	Lot # 10251935
<b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in CutSmart® 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
<b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in Antarctic Phosphatase Reaction Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> 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
<b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in NEBuffer 4 containing 1 µg 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	Pass

Assay Name/Specification	Lot # 10251935
electrophoresis.	
<b>Protein Purity Assay (SDS-PAGE)</b> Antarctic Phosphatase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	<b>Pass</b>
<b>RNase Activity (Extended Digestion)</b> 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.	<b>Pass</b>
<b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 5 units of Antarctic Phosphatase 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.	<b>Pass</b>

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

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Nicole Castagnozzi  
Production Scientist  
21 May 2024



Michael Tonello  
Packaging Quality Control Inspector  
31 Jul 2024