

## New England Biolabs Certificate of Analysis

**Product Name:** Antarctic Phosphatase  
**Catalog Number:** M0289L  
**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:** 10068198  
**Expiration Date:** 09/2021  
**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
M0289LVIAL	Antarctic Phosphatase	10052531	Pass
B0289SVIAL	Antarctic Phosphatase Reaction Buffer	10036011	Pass

Assay Name/Specification	Lot # 10068198
<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.	Pass
<b>qPCR DNA Contamination (E. coli Genomic)</b> 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
<b>Protein Purity Assay (SDS-PAGE)</b> Antarctic Phosphatase is ≥ 95% pure as determined by SDS-PAGE analysis using	Pass

Assay Name/Specification	Lot # 10068198
Coomassie Blue detection.	
<p><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 electrophoresis.</p>	<b>Pass</b>
<p><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] E. coli DNA and a minimum of 50 units of Antarctic Phosphatase incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	<b>Pass</b>
<p><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 &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	<b>Pass</b>

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



Ana Egana  
Production Scientist  
13 Mar 2020



Michael Tonello  
Packaging Quality Control Inspector  
13 Mar 2020