

## New England Biolabs Certificate of Analysis

**Product Name:** *phi29 DNA Polymerase*  
**Catalog Number:** *M0269L*  
**Concentration:** *10,000 U/ml*  
**Unit Definition:** *One unit is defined as the amount of enzyme that will incorporate 0.5 pmol of dNTP into acid insoluble material in 10 minutes at 30°C.*  
**Packaging Lot Number:** *10065461*  
**Expiration Date:** *10/2021*  
**Storage Temperature:** *-20°C*  
**Storage Conditions:** *10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 0.5 % Tween® 20 , 0.5 % IGEPAL® CA-630 , 50 % Glycerol, (pH 7.4 @ 25°C)*  
**Specification Version:** *PS-M0269S/L v3.0*

phi29 DNA Polymerase Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0269LVIAL	phi29 DNA Polymerase	10064397	Pass
B9000SVIAL	BSA, Molecular Biology Grade	10057616	Pass
B0269SVIAL	Φ29 DNA Polymerase Reaction Buffer	10064333	Pass


Assay Name/Specification	Lot # 10065461
<b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 10 units of phi29 DNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> phi29 DNA Polymerase is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
<b>Phosphatase Activity (pNPP)</b> A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl <sub>2</sub> containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units phi29 DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>RNase Activity Assay</b> A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA	Pass

Assay Name/Specification	Lot # 10065461
<p>and a minimum of 1 µl of phi29 DNA Polymerase is incubated at 37°C. After incubation for 4 hours, the substrate RNA is assessed by gel electrophoresis using fluorescent detection and compared to the product's RNase QC Standard resulting in no additional non-specific nuclease degradation.</p>	
<p><b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 10 units of phi29 DNA Polymerase 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.</p>	<b>Pass</b>
<p><b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in NEBuffer 2 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 units of phi29 DNA Polymerase 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.



Ben Penta  
Production Scientist  
04 Nov 2019



Jay Minichiello  
Packaging Quality Control Inspector  
20 Mar 2020