

## 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:** *10243053*  
**Expiration Date:** *04/2026*  
**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	10238463	Pass
B9200SVIAL	Recombinant Albumin, Molecular Biology G	10198642	Pass
B0269SVIAL	Φ29 DNA Polymerase Reaction Buffer	10233855	Pass

Assay Name/Specification	Lot # 10243053
<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 <10% conversion to the nicked form as determined by agarose gel electrophoresis.	<b>Pass</b>
<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.	<b>Pass</b>
<b>Phosphatase Activity (pNPP)</b> A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 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.	<b>Pass</b>
<b>Protein Purity Assay (SDS-PAGE)</b>	<b>Pass</b>

Assay Name/Specification	Lot # 10243053
<p>phi29 DNA Polymerase is <math>\geq 99\%</math> pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<p><b>Pass</b></p>
<p><b>RNase Activity Assay</b> A 10 <math>\mu</math>l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 <math>\mu</math>l 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 <math>\leq 1</math> E. coli genome.</p>	

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

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