

## New England Biolabs Certificate of Analysis

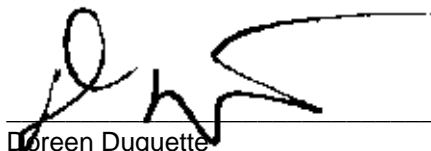
**Product Name:** *phi29 DNA Polymerase*  
**Catalog Number:** *M0269L*  
**Concentration:** *10,000 U/ml*  
**Unit Definition:** *One unit is defined as the amount of enzyme that will incorporate 0.5 pmol of dNTP into acid insoluble material in 10 minutes at 30°C.*  
**Packaging Lot Number:** *10061055*  
**Expiration Date:** *08/2021*  
**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               | 10049719   | Pass                 |
| B9000SVIAL                          | BSA, Molecular Biology Grade       | 10057616   | Pass                 |
| B0269SVIAL                          | Φ29 DNA Polymerase Reaction Buffer | 10049723   | Pass                 |

| Assay Name/Specification   | Lot # 10061055 |
|--|----------------|
| <p><b>Non-Specific DNase Activity (16 Hour)</b><br/>           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.</p>                                      | Pass           |
| <p><b>qPCR DNA Contamination (E. coli Genomic)</b><br/>           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> | Pass           |
| <p><b>Endonuclease Activity (Nicking)</b><br/>           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>   | Pass           |

| Assay Name/Specification  | Lot # 10061055 |
|---|----------------|
| <p><b>RNase Activity Assay</b><br/>A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA 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> | <b>Pass</b>    |
| <p><b>Protein Purity Assay (SDS-PAGE)</b><br/>phi29 DNA Polymerase is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>   | <b>Pass</b>    |
| <p><b>Phosphatase Activity (pNPP)</b><br/>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 &lt;0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.</p>  | <b>Pass</b>    |

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



Green Duquette  
Production Scientist  
13 Aug 2019



Michael Tonello  
Packaging Quality Control Inspector  
12 Dec 2019