

New England Biolabs Certificate of Analysis

Product Name: *phi29 DNA Polymerase*
Catalog Number: *M0269S*
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: *10113084*
Expiration Date: *04/2023*
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 |

| Assay Name/Specification | Lot # 10113084 |
|--|----------------|
| <p>RNase Activity Assay 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> | Pass |
| <p>Non-Specific DNase Activity (16 Hour) 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>Phosphatase Activity (pNPP) 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.</p> | Pass |
| <p>qPCR DNA Contamination (E. coli Genomic) 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</p> | Pass |

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|---|----------------|
| <p>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> | |
| <p>Protein Purity Assay (SDS-PAGE) phi29 DNA Polymerase is $\geq 99\%$ pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p> | Pass |
| <p>Endonuclease Activity (Nicking) 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.</p> | Pass |

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.



Christie Vazquez
Production Scientist
11 Jun 2021



Josh Hersey
Packaging Quality Control Inspector
11 Jun 2021