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240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## 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:	10160065
Expiration Date:	05/2024
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	10151191	Pass	
B9200SVIAL	Recombinant Albumin, Molecular Biology G	10150376	Pass	
B0269SVIAL	Φ29 DNA Polymerase Reaction Buffer	10064333	Pass	

Assay Name/Specification	Lot # 10160065
Protein Purity Assay (SDS-PAGE) phi29 DNA Polymerase is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
<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 $\leq$ 1 E. coli genome.	Pass
<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.	Pass
Endonuclease Activity (Nicking)	Pass





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Assay Name/Specification	Lot # 10160065
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>RNase Activity Assay</b> A 10 $\mu$ I reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 $\mu$ I 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.	Pass
<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

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.

vistie Vazquez

Christie Vazquez Production Scientist 27 Jul 2022

Erin Varney

Packaging Quality Control Inspector 27 Jul 2022

