

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:	Φ29 DNA Polymerase Reaction Buffer
Catalog #:	B0269S
Concentration:	10X Concentrate
Lot #:	0021706
Assay Date:	06/2017
Expiration Date:	6/2022
Storage Temp:	-20°C
Composition (1X):	50 mM Tris-HCl, 10 mM MgCl2, 10 mM (NH4)2 SO4, 4 mM DTT, (pH 7.5 @ 25°C)
Specification Version:	PS-B0269S v1.0
Effective Date:	10 May 2017

Assay Name/Specification (minimum release criteria)	Lot #0021706
<b>Endonuclease Activity (Nicking, Buffer)</b> - A 50 $\mu$ l reaction in 2X phi29 DNA Polymerase Reaction Buffer containing 1 $\mu$ g of supercoiled PhiX174 DNA incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Non-Specific DNase Activity (16 hour, Buffer)</b> - A 50 $\mu$ l reaction in 2X phi29 DNA Polymerase Reaction Buffer containing 1 $\mu$ g of T3 DNA in addition to a reaction containing Lambda-HindIII DNA 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>pH (buffers/solutions)</b> - The pH of 10X phi29 DNA Polymerase Reaction Buffer is between pH 7.4 and 7.6 at 25°C.	Pass
<b>Phosphatase Activity (pNPP, Buffer)</b> - A 200 $\mu$ l reaction in 1M Diethanolamine @ pH 9.8 and 0.5 mM MgCl <sub>2</sub> containing 2.5 mM <i>p</i> -Nitrophenyl Phosphate (pNPP) and a minimum of 40 $\mu$ l phi29 DNA Polymerase Reaction Buffer incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>qPCR DNA Contamination (</b> <i>E. coli</i> <b>Genomic)</b> - A minimum of 1 $\mu$ l of phi29 DNA Polymerase Reaction Buffer is screened for the presence of <i>E. coli</i> genomic DNA using SYBR® Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is $\leq 1$ <i>E. coli</i> genome.	Pass



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Assay Name/Specification (minimum release criteria)	Lot #0021706
<b>RNase Activity (Extended Digestion)</b> - A 10 $\mu$ l reaction in NEBuffer 4 containing 40 ng of a 300 base single- stranded RNA and a minimum of 1 $\mu$ l of phi29 DNA Polymerase Reaction Buffer is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass

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Authorized by Karen Moreira 10 May 2017



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Inspected by Tony Spear-Alfonso 01 Jun 2017