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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:	Q5® Reaction Buffer Pack
Catalog Number:	B9027S
Concentration:	5 X Concentrate
Packaging Lot Number:	10173754
Expiration Date:	07/2025
Storage Temperature:	-20°C
Specification Version:	PS-B9027S v2.0
Composition (1X):	Proprietary

Q5® Reaction Buffer Pack Component List			
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result
B9028AVIAL	Q5® High GC Enhancer	10165337	Pass
B9027SVIAL	Q5® Reaction Buffer Pack	10165336	Pass

Assay Name/Specification	Lot # 10173754
<b>qPCR DNA Contamination (E. coli Genomic, Buffer)</b> A minimum of 1 $\mu$ l of Q5® Reaction Buffer 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
PCR Amplification (7 kb Human Genomic DNA, Buffer) A 50 $\mu$ I reaction in Q5® Reaction Buffer in the presence of 200 $\mu$ M dNTPs and 0.5 $\mu$ M primers containing 20 ng Human Genomic DNA with 1 unit of Q5® High-Fidelity DNA Polymerase for 30 cycles of PCR amplification results in the expected 7 kb product.	Pass
PCR Amplification (20 kb Lambda DNA, Buffer) A 50 $\mu$ I reaction in Q5 <sup>®</sup> Reaction Buffer in the presence of 200 $\mu$ M dNTPs and 1 $\mu$ M primers containing 10 ng Lambda DNA with 1 unit of Q5 <sup>®</sup> High-Fidelity DNA Polymerase for 22 cycles of PCR amplification results in the expected 20 kb product.	Pass
<b>Phosphatase Activity (pNPP, Buffer)</b> A 200 µl reaction in 1M Diethanolamine @ pH 9.8 and 0.5 mM MgCl2 containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 80 µl Q5® Reaction Buffer incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass





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Assay Name/Specification	Lot # 10173754	
<b>RNAse Activity Assay (4 Hour 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 Q5® Reaction Buffer is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass	
Endonuclease Activity (Nicking, Buffer) A 50 $\mu$ I reaction in 2X Q5® 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 µl reaction in 2X Q5® Reaction Buffer containing 1 µg of T3 or T7 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	

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

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Lea Antonpoulos Production Scientist 29 Sep 2022

Much

Michael Tonello Packaging Quality Control Inspector 09 Jan 2023

