

## New England Biolabs Certificate of Analysis

Product Name: Q5<sup>®</sup> Reaction Buffer Pack  
 Catalog Number: B9027S  
 Concentration: 5 X Concentrate  
 Packaging Lot Number: 10138670  
 Expiration Date: 06/2024  
 Storage Temperature: -20°C  
 Specification Version: PS-B9027S v2.0  
 Composition (1X): Proprietary

Q5 <sup>®</sup> Reaction Buffer Pack Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
B9028AVIAL	Q5 <sup>®</sup> High GC Enhancer	10111977	Pass
B9027SVIAL	Q5 <sup>®</sup> Reaction Buffer Pack	10111976	Pass

Assay Name/Specification	Lot # 10138670
<p><b>PCR Amplification (7 kb Human Genomic DNA, Buffer)</b>            A 50 µl reaction in Q5<sup>®</sup> Reaction Buffer in the presence of 200 µM dNTPs and 0.5 µM primers containing 20 ng Human Genomic DNA with 1 unit of Q5<sup>®</sup> High-Fidelity DNA Polymerase for 30 cycles of PCR amplification results in the expected 7 kb product.</p>	Pass
<p><b>PCR Amplification (20 kb Lambda DNA, Buffer)</b>            A 50 µl reaction in Q5<sup>®</sup> Reaction Buffer in the presence of 200 µM dNTPs and 1 µM 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.</p>	Pass
<p><b>Non-Specific DNase Activity (16 hour, Buffer)</b>            A 50 µl reaction in 2X Q5<sup>®</sup> 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.</p>	Pass
<p><b>qPCR DNA Contamination (E. coli Genomic, Buffer)</b>            A minimum of 1 µl of Q5<sup>®</sup> Reaction Buffer is screened for the presence of E. coli genomic DNA using SYBR<sup>®</sup> 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

Assay Name/Specification	Lot # 10138670
<p><b>RNase Activity Assay (4 Hour Digestion)</b> A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Q5<sup>®</sup> Reaction Buffer is incubated at 37°C. After incubation for 4 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>	<b>Pass</b>
<p><b>Phosphatase Activity (pNPP, Buffer)</b> A 200 µl reaction in 1M Diethanolamine @ pH 9.8 and 0.5 mM MgCl<sub>2</sub> containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 80 µl Q5<sup>®</sup> Reaction Buffer 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>
<p><b>Endonuclease Activity (Nicking, Buffer)</b> A 50 µl reaction in 2X Q5<sup>®</sup> Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	<b>Pass</b>

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

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15 Feb 2022



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