

## New England Biolabs Certificate of Analysis

**Product Name:** Q5® Reaction Buffer Pack  
**Catalog Number:** B9027S  
**Concentration:** 5 X Concentrate  
**Lot Number:** 10035289  
**Expiration Date:** 10/2021  
**Storage Temperature:** -20°C  
**Specification Version:** PS-B9027S v1.0  
**Composition (1X):** Proprietary

Q5® Reaction Buffer Pack Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
B9028AVIAL	Q5® High GC Enhancer	10034782	Pass
B9027SVIAL	Q5® Reaction Buffer Pack	10023544	Pass

Assay Name/Specification	Lot # 10035289
<p><b>PCR Amplification (7 kb Human Genomic DNA, Buffer)</b>            A 50 µl reaction in Q5® 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® High-Fidelity DNA Polymerase for 30 cycles of PCR amplification results in the expected 7 kb product.</p>	Pass
<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® Reaction Buffer incubated for 4 hours at 37°C yields &lt;0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.</p>	Pass
<p><b>qPCR DNA Contamination (E. coli Genomic, Buffer)</b>            A minimum of 1 µl 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 ≤ 1 E. coli genome.</p>	Pass
<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® Reaction Buffer is incubated at 37°C. After incubation for 4 hours, &gt;90% of the substrate RNA remains intact as determined by gel</p>	Pass

Assay Name/Specification	Lot # 10035289
<p>electrophoresis using fluorescent detection.</p> <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 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>	<b>Pass</b>
<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>	<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.



Tony Spear-Alfonso  
Production Scientist  
05 Nov 2018



Josh Hersey  
Packaging Quality Control Inspector  
21 Mar 2019