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New England Biolabs Certificate of Analysis

Product Name: Q5® Reaction Buffer Pack

Catalog Number: B9027S

Concentration: 5 X Concentrate

Packaging Lot Number: 10156966
Expiration Date: 03/2025
Storage Temperature: -20°C

Specification Version: PS-B9027S v2.0 Composition (1X): Proprietary

| Q5® Reaction Buffer Pack Component List | | | | |
|---|--------------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| B9028AVIAL | Q5® High GC Enhancer | 10140762 | Pass | |
| B9027SVIAL | Q5® Reaction Buffer Pack | 10151185 | Pass | |

| Assay Name/Specification | Lot # 10156966 |
|---|----------------|
| Non-Specific DNase Activity (16 hour, Buffer) 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 |
| RNAse Activity Assay (4 Hour Digestion) 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, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection. | Pass |
| qPCR DNA Contamination (E. coli Genomic, Buffer) 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. | Pass |
| Phosphatase Activity (pNPP, Buffer) 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 | Pass |



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| Assay Name/Specification | Lot # 10156966 | |
|---|----------------|--|
| for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as | | |
| determined by spectrophotometric analysis. | | |
| PCR Amplification (20 kb Lambda DNA, Buffer) | Pass | |
| A 50 μI reaction in Q5® Reaction Buffer in the presence of 200 μM dNTPs and 1 μM | | |
| primers containing 10 ng Lambda DNA with 1 unit of Q5® High-Fidelity DNA Polymerase | | |
| for 22 cycles of PCR amplification results in the expected 20 kb product. | | |
| PCR Amplification (7 kb Human Genomic DNA, Buffer) | Pass | |
| 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. | | |
| Endonuclease Activity (Nicking, Buffer) | Pass | |
| A 50 µl reaction in 2X Q5® Reaction Buffer containing 1 µg 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. | | |

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.

Christie Vazquez Production Scientist 19 Jul 2022

histie Vanguez

Michael Tonello

Packaging Quality Control Inspector

19 Jul 2022



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