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## New England Biolabs Certificate of Analysis

Product Name: Q5® High-Fidelity 2X Master Mix

Catalog Number: M0492S

Concentration: 2 X Concentrate

Packaging Lot Number: 10170531
Expiration Date: 10/2024
Storage Temperature: -20°C

Specification Version: PS-M0492S/L v2.0

Composition (1X): Proprietary

| Q5® High-Fidelity 2X Master Mix Component List |                                 |            |                      |  |
|--|---------------------------------|------------|----------------------|--|
| <b>NEB Part Number</b>                         | Component Description           | Lot Number | Individual QC Result |  |
| M0492SVIAL                                     | Q5® High-Fidelity 2X Master Mix | 10168019   | Pass                 |  |

| Assay Name/Specification  | Lot # 10170531 |
|---|----------------|
| Protein Purity Assay (SDS-PAGE) Q5® High-Fidelity DNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.   | Pass           |
| <b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 2 units of Q5® High-Fidelity DNA Polymerase 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           |
| RNase Activity (Extended 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® High-Fidelity 2X Master Mix 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           |
| PCR Amplification (7 kb Human Genomic DNA, Master Mix) A 50 μl reaction in 1X Q5® High-Fidelity Master Mix and 0.5 μM primers containing 20 ng Human Genomic DNA for 30 cycles of PCR amplification results in the expected 7 kb product.   | Pass           |
| PCR Amplification (20 kb Lambda DNA, Master Mix)  | Pass           |



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| Assay Name/Specification  | Lot # 10170531 |
|---|----------------|
| A 50 $\mu$ l reaction in 1X Q5® High-Fidelity Master Mix and 1.0 $\mu$ M primers containing 10 ng Lambda DNA for 22 cycles of PCR amplification results in the expected 20 kb product.  |                |
| Phosphatase Activity (pNPP) A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of Q5® High-Fidelity DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.         | Pass           |
| Endonuclease Activity (Nicking, Polymerase, dNTP) A 50 μl reaction in NEBuffer 2 in the presence of 400 μM dNTPs containing 1 μg of supercoiled pUC19 DNA and a minimum of 10 units of Q5® High-Fidelity DNA Polymerase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass           |
| Non-Specific DNase Activity (16 hour, Buffer) A 50 µl reaction in 1X Q5® High-Fidelity Master Mix 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.

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.

Lea Antonpoulos

Production Scienti

07 Nov 2022

Michael Tonello

Packaging Quality Control Inspector

29 Nov 2022



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