

New England Biolabs Certificate of Analysis


Product Name: Q5[®] Hot Start High-Fidelity 2X Master Mix
Catalog #: M0494S/L
Concentration: 2X
Lot #: 0251709
Assay Date: 09/2017
Expiration Date: 09/2019
Storage Temp: -20°C
Composition (1X): Proprietary
Specification Version: PS-M0494S/L v1.0
Effective Date: 15 Sep 2017

Assay Name/Specification (minimum release criteria)	Lot #0251709
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 [®] Hot Start High-Fidelity Master Mix 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.	Pass
PCR Amplification (20 kb Lambda DNA, Master Mix) - A 50 µl reaction in 1X Q5 [®] Hot Start High-Fidelity Master Mix and 1.0 µM primers containing 10 ng Lambda DNA for 22 cycles of PCR amplification results in the expected 20 kb product.	Pass
PCR Amplification (7 kb Human Genomic DNA, Master Mix) - A 50 µl reaction in 1X Q5 [®] Hot Start 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 (Hot Start, Human Genomic DNA, Master Mix) - A 25 µl reaction in 1X Q5 [®] Hot Start High-Fidelity Master Mix and 0.5 µM primers containing 50 ng Human Genomic DNA for 25 cycles of PCR amplification results in the expected 665 bp product and a decrease in non-specific genomic bands after pre-incubation at room temperature for 1 hour, when compared to a non-hot start control reaction.	Pass

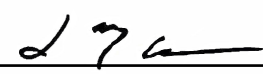


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Assay Name/Specification (minimum release criteria)	Lot #0251709
<p>Phosphatase Activity (pNPP) - A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl₂ containing 2.5 mM <i>p</i>-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of Q5[®] High-Fidelity DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.</p>	Pass
<p>Protein Purity Assay (SDS-PAGE) - Q5[®] High-Fidelity DNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	Pass
<p>qPCR DNA Contamination (<i>E. coli</i> Genomic) - A minimum of 2 units of Q5[®] High-Fidelity DNA Polymerase is screened for the presence of <i>E. coli</i> genomic DNA using SYBR[®] Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is ≤ 1 <i>E. coli</i> genome.</p>	Pass
<p>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[®] Hot Start 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.</p>	Pass



 Authorized by
 Melanie Fortier
 15 Sep 2017



 Inspected by
 Lynne Apone
 09 Nov 2017

