

New England Biolabs Product Specification

Product Name:	<i>Taq 5X Master Mix</i>
Catalog #:	<i>M0285S/L</i>
Concentration:	<i>5X Concentrate</i>
Shelf Life:	<i>24 months</i>
Storage Temp:	<i>-20°C</i>
Composition (1X):	<i>10 mM Tris-HCl (pH 8.6 @ 25°C), 50 mM KCl, 1.5 mM MgCl₂, 0.2 mM dATP, 0.2 mM dCTP, 0.2 mM dGTP, 0.2 mM dTTP, 5 % Glycerol, 0.08 % IGEPAL® CA-630, 0.05 % Tween® 20, 25 units/ml Taq DNA Polymerase</i>
Specification Version:	<i>PS-M0285S/L v3.0</i>
Effective Date:	<i>31 Jul 2024</i>

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of *Taq* DNA Polymerase incubated for 4 hours at 37°C and 75°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Non-Specific DNase Activity (16 hour, Buffer) - A 50 µl reaction in 2X *Taq* 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.

PCR Amplification (5 kb Lambda, Master Mix) - A 25 µl reaction in 1X *Taq* Master Mix and 0.2 µM primers containing 5 ng Lambda DNA for 25 cycles of PCR amplification results in the expected 5 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 *Taq* DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.

Protein Purity (Microfluidic Electrophoresis) - *Taq* DNA Polymerase is ≥97% pure as determined by microfluidic electrophoresis.

qPCR DNA Contamination (*E. coli* Genomic) - A minimum of 5 units of *Taq* 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.

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 *Taq* 5X 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.

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Assay Name/Specification (minimum release criteria)

Single Stranded DNase Activity (FAM-Labeled Oligo) - A 50 µl reaction in ThermoPol® Reaction Buffer containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 25 units of <i>Taq</i> DNA Polymerase incubated for 30 minutes at 37°C and 75°C yields <10% degradation as determined by capillary electrophoresis.
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Date 31 Jul 2024

Lauren Brown
Quality Approver

