

New England Biolabs Certificate of Analysis

Product Name: Hot Start Taq DNA Polymerase
Catalog Number: M0495S
Concentration: 5,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme that will incorporate 15 nmol of dNTP into acid insoluble material in 30 minutes at 75°C.
Lot Number: 10015904
Expiration Date: 08/2020
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 1X Stabilizers, 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version: PS-M0495S/L v1.0

Hot Start Taq DNA Polymerase Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0495SVIAL	Hot Start Taq DNA Polymerase	10019649	Pass
B9014SVIAL	Standard Taq Reaction Buffer Pack	10022003	Pass

Assay Name/Specification	Lot # 10015904
<p>qPCR DNA Contamination (E. coli Genomic) A minimum of 5 units of Hot Start 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.</p>	Pass
<p>Protein Purity Assay (SDS-PAGE) Taq DNA Polymerase is $\geq 99\%$ pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</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 Hot Start Taq DNA Polymerase is incubated at 37°C. After incubation for 16 hours, $>90\%$ of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>	Pass
<p>Single Stranded DNase Activity (Hot Start, FAM-Labeled Oligo) A 50 μl reaction in ThermoPol® Reaction Buffer containing a 10 nM solution of a</p>	Pass

Assay Name/Specification	Lot # 10015904
<p>fluorescent internal labeled oligonucleotide and a minimum of 25 units of Taq DNA Polymerase incubated for 30 minutes at either 37°C or 75°C yields <10% degradation as determined by capillary electrophoresis.</p>	
<p>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 Taq 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>PCR Amplification (Hot Start 2 kb Lambda DNA) A 50 µl reaction in ThermoPol[®] Reaction Buffer in the presence of 200 µM dNTPs and 0.2 µM primers containing 20 pg Lambda DNA and 100 ng Human Genomic DNA with 1.25 units of Hot Start Taq DNA Polymerase for 30 cycles of PCR amplification results in an increase in yield of the 2 kb Lambda product and a decrease in non-specific genomic bands when compared to a non-hot start control reaction.</p>	Pass
<p>PCR Amplification (5.0 kb Lambda DNA) A 50 µl reaction in ThermoPol[®] Reaction Buffer in the presence of 200 µM dNTPs and 0.2 µM primers containing 5 ng Lambda DNA with 1.25 units of Hot Start Taq DNA Polymerase for 25 cycles of PCR amplification results in the expected 5.0 kb product.</p>	Pass
<p>Non-Specific DNase Activity (16 Hour) A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 5 units of Hot Start Taq DNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	Pass
<p>Inhibition of Primer Extension (Hot Start, Radioactivity Incorporation) A 50 µl primer extension assay in ThermoPol[®] Reaction Buffer in the presence of 200 µM dNTPs including [³H]-dTTP, containing 15 nM primed single-stranded M13mp18 with 2.5 units of Hot Start Taq DNA Polymerase incubated for 16 hours at 25°C yields >95% inhibition when compared to a non-hot start control reaction.</p>	Pass
<p>Endonuclease Activity (Nicking, Hot Start) 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 either 37°C or 75°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass

This product has been tested and shown to be in compliance with all specifications.



Tony Spear-Alfonso
Production Scientist
27 Aug 2018



Michael Tonello
Packaging Quality Control Inspector
12 Oct 2018