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New England Biolabs Certificate of Analysis

Product Name: OneTag® Hot Start DNA Polymerase

Catalog Number: M0481S 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.

Packaging Lot Number: 10057355
Expiration Date: 07/2021
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 0.5 % Tween®

20 , 0.5 % IGEPAL® CA-630 , 50 % Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-M0481S/L/X v1.0

| OneTaq® Hot Start DNA Polymerase Component List | | | | |
|---|----------------------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| M0481SVIAL | OneTaq® Hot Start DNA Polymerase | 10049407 | Pass | |
| B9026AVIAL | OneTaq® High GC Enhancer | 10031487 | Pass | |
| B9023SVIAL | OneTaq® GC Reaction Buffer | 10034012 | Pass | |
| B9022SVIAL | OneTaq® Standard Reaction Buffer | 10034011 | Pass | |

| Assay Name/Specification | Lot # 10057355 |
|--|----------------|
| 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 OneTaq® Hot Start DNA Polymerase incubated for 16 hours at 25°C yields >95% inhibition when compared to a non-hot start control reaction. | Pass |
| 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 OneTaq® Hot Start 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. | Pass |
| PCR Amplification (5.0 kb Lambda DNA) A 25 μl reaction in OneTaq® Standard Reaction Buffer in the presence of 200 μM dNTPs and 0.2 μM primers containing 5 ng Lambda DNA with 0.625 units of OneTaq® Hot Start DNA Polymerase for 25 cycles of PCR amplification results in the expected 5.0 kb product. | Pass |



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| Assay Name/Specification | Lot # 10057355 |
|--|----------------|
| PCR Amplification (Buffer Dependent, >65% GC-rich) A 25 μl reaction in OneTaq® GC Buffer in the presence of 200 μM dNTPs and 0.2 μM primers containing 10 ng Human Genomic DNA with 0.625 units of OneTaq® Hot Start DNA Polymerase for 30 cycles of PCR amplification results in the buffer-dependent production of the expected 737 bp product. | Pass |
| PCR Amplification (Enhancer Dependent, >70% GC-rich) A 25 µl reaction in OneTaq® GC Reaction Buffer and 20% OneTaq® High GC Enhancer in the presence of 200 µM dNTPs and 0.2 µM primers containing 10 ng Human Genomic DNA with 0.625 units of OneTaq® Hot Start DNA Polymerase for 30 cycles of PCR amplification results in the enhancer-dependent production of the expected 627 bp product. | Pass |
| PCR Amplification (Hot Start 2 kb Lambda DNA) A 25 μl reaction in OneTaq® Standard Reaction Buffer in the presence of 200 μM dNTPs and 0.2 μM primers containing 10 pg Lambda DNA and 50 ng Human Genomic DNA with 0.625 units of OneTaq® Hot Start 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. | 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 OneTaq® Hot Start 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. | Pass |

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

Derek Robinson **Production Scientist**

31 Jul 2019

Michael Tonello

Packaging Quality Control Inspector 20 Nov 2019



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