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New England Biolabs Certificate of Analysis

Product Name: OneTag® Hot Start Quick-Load® 2X Master Mix with GC Buffer

Catalog Number: M0489S

Concentration: 2 X Concentrate

Packaging Lot Number: 10206181
Expiration Date: 07/2025
Storage Temperature: -20°C

Specification Version: PS-M0489S/L v2.0

Composition (1X): 80 mM Tris-SO4 (pH 9.2 @ 25°C), 20 mM (NH4)2SO4, 2 mM MgSO4, 0.2 mM

dATP, 0.2 mM dCTP, 0.2 mM dGTP, 0.2 mM dTTP, 5 % Glycerol, 5 % DMSO,

0.06 % IGEPAL® CA-630, 0.05 % Tween® 20, 1 X Xylene cyanol, 1 X

Tartrazine, 25 units/ml OneTaq® Hot Start DNA Polymerase

| OneTaq® Hot Start Quick-Load® 2X Master Mix with GC Buffer Component List | | | | |
|---|--|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| M0489SVIAL | OneTaq® Hot Start Quick-Load® 2X Master Mix with GC Buffer | 10202622 | Pass | |
| B9026AVIAL | OneTaq® High GC Enhancer | 10179724 | Pass | |

| Assay Name/Specification | Lot # 10206181 |
|--|----------------|
| 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, Buffer) A 50 µl reaction in 1X OneTaq® Hot Start Quick-Load® Master Mix with GC Buffer 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 |
| PCR Amplification (Buffer Dependent, >65% GC-rich, Master Mix) A 25 μl reaction in 1X OneTaq® Hot Start Quick-Load® Master Mix with GC Buffer and 0.2 μM primers containing 10 ng Human Genomic DNA for 30 cycles of PCR amplification results in the buffer-dependent production of the 737 bp product. | Pass |
| PCR Amplification (Enhancer Dependent, >70% GC-rich, Master Mix) | Pass |



M0489S / Lot: 10206181

Page 1 of 2

| Assay Name/Specification | Lot # 10206181 |
|--|----------------|
| A 25 µl reaction in 1X OneTaq® Hot Start Quick-Load® Master Mix with GC Buffer and 20% OneTaq® High GC Enhancer in the presence of 0.2 µM primers containing 10 ng Human Genomic DNA for 30 cycles of PCR amplification results in the enhancer-dependent production of the 627 bp product. | |
| 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 Quick-Load® 2X Master Mix with GC Buffer 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 |

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 Scientist

15 Mar 2023

Josh Hersey

Packaging Quality Control Inspector

27 Sep 2023



M0489S / Lot: 10206181

Page 2 of 2