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## New England Biolabs Certificate of Analysis

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

Catalog Number: M0485S

Concentration: 2 X Concentrate

Packaging Lot Number: 10175065
Expiration Date: 12/2024
Storage Temperature: -20°C

Specification Version: PS-M0485S/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, 25 units/ml OneTag® Hot

Start DNA Polymerase

OneTag® Hot Start 2X Master Mix with GC Buffer Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
M0485SVIAL	OneTaq® Hot Start 2X Master Mix with GC Buffer	10174433	Pass	
B9026AVIAL	OneTaq® High GC Enhancer	10157599	Pass	

Assay Name/Specification	Lot # 10175065
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 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 (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



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Assay Name/Specification	Lot # 10175065
PCR Amplification (Enhancer Dependent, >70% GC-rich, Master Mix) A 25 µl reaction in 1X OneTaq® Hot Start 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.	Pass
PCR Amplification (Buffer Dependent, >65% GC-rich, Master Mix) A 25 μl reaction in 1X OneTaq® Hot Start 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
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 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

04 Jan 2023**¢** 

Michael Tonello

Packaging Quality Control Inspector

09 Jan 2023

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