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New England Biolabs Certificate of Analysis

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

Catalog Number: M0484L

Concentration: 2 X Concentrate

Packaging Lot Number: 10221155
Expiration Date: 11/2025
Storage Temperature: -20°C

Specification Version: PS-M0484S/L v2.0

Composition (1X): 20 mM Tris-HCl (pH 8.9 @ 25°C), 22 mM NH4Cl, 22 mM KCl, 1.8 mM

MgCl2, 0.2 mM dATP, 0.2 mM dCTP, 0.2 mM dGTP, 0.2 mM dTTP, 5 % Glycerol, 0.06 % IGEPAL® CA-630, 0.05 % Tween® 20, 25 units/ml

OneTaq® Hot Start DNA Polymerase

OneTaq® Hot Start 2X Master Mix with Standard Buffer Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
M0484SVIAL	OneTaq® Hot Start 2X Master Mix with Standard Buffer	10221157	Pass	

Assay Name/Specification	Lot # 10221155
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 Standard 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 (5 kb Lambda, Master Mix) A 25 μl reaction in 1X OneTaq® Hot Start Master Mix with Standard Buffer and 0.2 μM primers containing 5 ng Lambda DNA for 25 cycles of PCR amplification results in the expected 5 kb 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	Pass



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Assay Name/Specification	Lot # 10221155
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.	
RNase Activity (Extended Digestion)	Pass
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 Standard Buffer is	
incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains	

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

intact as determined by gel electrophoresis using fluorescent detection.

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 | Reduction Scientist

20 Jan 2024

Michael Tonello

Packaging Quality Control Inspector

22 Jan 2024



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