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## New England Biolabs Certificate of Analysis

Product Name: LongAmp® Hot Start Tag 2X Master Mix

Catalog Number: M0533L

Concentration: 2 X Concentrate

Packaging Lot Number: 10061098
Expiration Date: 05/2021
Storage Temperature: -20°C

Specification Version: PS-M0533S/L v1.0

Composition (1X): 60 mM Tris-SO4 (pH 9.1 @ 25°C), 20 mM (NH4)2SO4, 2 mM MgSO4, 0.3 mM

dATP, 0.3 mM dCTP, 0.3 mM dGTP, 0.3 mM dTTP, 3 % Glycerol, 0.06 % IGEPAL® CA-630, 0.05 % Tween® 20, 125 units/ml LongAmp® Hot Start

Taq DNA Polymerase

LongAmp® Hot Start Taq 2X Master Mix Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
M0533SVIAL	LongAmp® Hot Start Taq 2X Master Mix	10057281	Pass	

Assay Name/Specification	Lot # 10061098
PCR Amplification (30 kb Human Genomic DNA, Master Mix) A 25 µl reaction in 1X LongAmp® Hot Start Taq Master Mix and 0.4 µM primers containing 500 ng Human Genomic DNA for 28 cycles of PCR amplification results in the expected 30 kb product.	Pass
PCR Amplification (30 kb Lambda DNA, Master Mix) A 25 µl reaction in 1X LongAmp® Hot Start Taq Master Mix and 0.4 µM primers containing 1 ng Lambda DNA for 28 cycles of PCR amplification results in the expected 30 kb product.	Pass
PCR Amplification (Hot Start, Human Genomic DNA, Master Mix) A 50 μl reaction in 1X LongAmp® Hot Start Taq Master Mix and 0.2 μM primers containing 2 ng Human Genomic DNA for 35 cycles of PCR amplification results in the expected 306 bp product and a decrease in non-specific genomic bands after pre-incubation at room temperature for 1 hour, when compared to a non-hot start control reaction.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 2.5 units of LongAmp® 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	Pass



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Assay Name/Specification	Lot # 10061098
purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	
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 LongAmp® Hot Start Taq 2X Master Mix 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
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 10 units of LongAmp® Hot Start Taq 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 LongAmp® Hot Start Taq Master Mix containing 1 µg of T3 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

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

Christie Vazquez Production Scientist 22 Nov 2019

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Michael Tonello

Packaging Quality Control Inspector

22 Nov 2019



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