NEW ENGLAND BioLabs Inc.
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New England Biolabs
Certificate of Analysis

Product Name:	*LongAmp® Hot Start Taq 2X Master Mix*
Catalog Number:	*M0533S*
Concentration:	*2 X Concentrate*
Packaging Lot Number:	*10134561*
Expiration Date:	*04/2023*
Storage Temperature:	*-20°C*
Specification Version:	*PS-M0533S/L v2.0*
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			
NEB Part Number	**Component Description**	**Lot Number**	**Individual QC Result**
M0533SVIAL	LongAmp® Hot Start Taq 2X Master Mix	10125136	**Pass**

Assay Name/Specification	**Lot # 10134561**
Non-Specific DNase Activity (16 hour, Buffer) A 50 µl reaction in 1X LongAmp® Hot Start Taq Master Mix containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA incubated for 16 hours at 37ºC results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	**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 LongAmp® Hot Start Taq 2X Master Mix is incubated at 37ºC. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	**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 purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	**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	**Pass**

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Assay Name/Specification	Lot # 10134561
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.	
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 [^{3}H]-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**
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 (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**

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.

Christie Vazquez
Production Scientist
13 Jan 2022

Michael Tonello
Packaging Quality Control Inspector
13 Jan 2022