

## New England Biolabs Certificate of Analysis

*Product Name:* OneTaq<sup>®</sup> Hot Start DNA Polymerase  
*Catalog #:* M0481S/L/X  
*Concentration:* 5,000 units/ml  
*Unit Definition:* One unit is defined as the amount of enzyme that will incorporate 15 nmol of dNTP into acid insoluble material in 30 minutes at 75°C.  
*Lot #:* 0101612  
*Assay Date:* 12/2016  
*Expiration Date:* 12/2018  
*Storage Temp:* -20°C  
*Storage Conditions:* 10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.5 % Tween<sup>®</sup> 20, 0.5 % IGEPAL<sup>®</sup> CA-630, 50 % Glycerol, (pH 7.4 @ 25°C)  
*Specification Version:* PS-M0481S/L/X v1.0  
*Effective Date:* 21 Apr 2016

Assay Name/Specification (minimum release criteria)	Lot #0101612
<p><b>Inhibition of Primer Extension (Hot Start, Radioactivity Incorporation)</b> - A 50 µl primer extension assay in ThermoPol<sup>®</sup> Reaction Buffer in the presence of 200 µM dNTPs including [<sup>3</sup>H]-dTTP, containing 15 nM primed single-stranded M13mp18 with 2.5 units of OneTaq<sup>®</sup> Hot Start DNA Polymerase incubated for 16 hours at 25°C yields &gt;95% inhibition when compared to a non-hot start control reaction.</p>	<b>Pass</b>
<p><b>Non-Specific DNase Activity (16 Hour)</b> - A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 5 units of OneTaq<sup>®</sup> Hot Start DNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	<b>Pass</b>
<p><b>PCR Amplification (5.0 kb Lambda DNA)</b> - A 25 µl reaction in OneTaq<sup>®</sup> Standard Reaction Buffer in the presence of 200 µM dNTPs and 0.2 µM primers containing 5 ng Lambda DNA with 0.625 units of OneTaq<sup>®</sup> Hot Start DNA Polymerase for 25 cycles of PCR amplification results in the expected 5.0 kb product.</p>	<b>Pass</b>
<p><b>PCR Amplification (Buffer Dependent, &gt;65% GC-rich)</b> - A 25 µl reaction in OneTaq<sup>®</sup> GC Buffer in the presence of 200 µM dNTPs and 0.2 µM primers containing 10 ng Human Genomic DNA with 0.625 units of OneTaq<sup>®</sup> Hot Start DNA Polymerase for 30 cycles of PCR amplification results in the buffer-dependent production of the expected 737 bp product.</p>	<b>Pass</b>

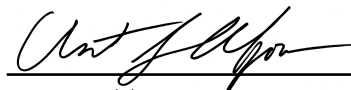


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Assay Name/Specification (minimum release criteria)	Lot #0101612
<p><b>PCR Amplification (Enhancer Dependent, &gt;70% GC-rich)</b> - A 25 µl reaction in OneTaq<sup>®</sup> GC Reaction Buffer and 20% OneTaq<sup>®</sup> High GC Enhancer in the presence of 200 µM dNTPs and 0.2 µM primers containing 10 ng Human Genomic DNA with 0.625 units of OneTaq<sup>®</sup> Hot Start DNA Polymerase for 30 cycles of PCR amplification results in the enhancer-dependent production of the expected 627 bp product.</p>	<b>Pass</b>
<p><b>PCR Amplification (Hot Start 2 kb Lambda DNA)</b> - A 25 µl reaction in OneTaq<sup>®</sup> 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<sup>®</sup> 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.</p>	<b>Pass</b>
<p><b>RNase Activity (Extended Digestion)</b> - 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<sup>®</sup> Hot Start DNA Polymerase is incubated at 37°C. After incubation for 16 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>	<b>Pass</b>



Authorized by  
Melanie Fortier  
21 Apr 2016



Inspected by  
Tony Spear-Alfonso  
28 Apr 2017

