

## New England Biolabs Certificate of Analysis

**Product Name:** *Sulfolobus DNA Polymerase IV*  
**Catalog #:** M0327S/L  
**Concentration:** 2,000 units/ml  
**Unit Definition:** One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid insoluble material in 30 minutes at 55°C.  
**Lot #:** 0011612  
**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, 50 % Glycerol, (pH 7.4 @ 25°C)  
**Specification Version:** PS-M0327S/L v1.0  
**Effective Date:** 17 May 2016

Assay Name/Specification (minimum release criteria)	Lot #0011612
<b>Endonuclease Activity (Nicking)</b> - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of <i>Sulfolobus</i> DNA Polymerase IV incubated for 4 hours at either 37°C or 55°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] <i>E. coli</i> DNA and a minimum of 20 units of <i>Sulfolobus</i> DNA Polymerase IV incubated for 4 hours at either 37°C or 55°C releases <0.1% of the total radioactivity.	<b>Pass</b>
<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 2 units of <i>Sulfolobus</i> DNA Polymerase IV incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>Protein Purity Assay (SDS-PAGE)</b> - <i>Sulfolobus</i> DNA Polymerase IV is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	<b>Pass</b>
<b>qPCR DNA Contamination (<i>E. coli</i> Genomic)</b> - A minimum of 2 units of <i>Sulfolobus</i> DNA Polymerase IV is screened for the presence of <i>E. coli</i> genomic DNA using SYBR® Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is ≤ 1 <i>E. coli</i> genome.	<b>Pass</b>

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Assay Name/Specification (minimum release criteria)	Lot #0011612
<b>Single Stranded DNase Activity (FAM-Labeled Oligo)</b> - A 20 µl reaction in ThermoPol <sup>®</sup> Reaction Buffer containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 20 units of <i>Sulfolobus</i> DNA Polymerase IV incubated for 30 minutes at either 37°C or 55°C yields <10% degradation as determined by capillary electrophoresis.	<b>Pass</b>



Authorized by  
Melanie Fortier  
17 May 2016



Inspected by  
Tony Spear-Alfonso  
02 Nov 2016

