

New England Biolabs Certificate of Analysis

Product Name: Phusion[®] Hot Start Flex DNA Polymerase
Catalog Number: M0535L
Concentration: 2,000 U/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 74°C.
Packaging Lot Number: 10140649
Expiration Date: 10/2023
Storage Temperature: -20°C
Storage Conditions: 20 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 200 µg/ml BSA , 1X Stabilizers , 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version: PS-M0535S/L v1.0

Phusion [®] Hot Start Flex DNA Polymerase Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0535LVIAL	Phusion [®] Hot Start Flex DNA Polymerase	10125138	Pass
B0519SVIAL	Phusion [®] GC Buffer Pack	10129313	Pass
B0518SVIAL	Phusion [®] HF Buffer Pack	10135556	Pass
B0515AVIAL	DMSO	10129312	Pass
B0510AVIAL	MgCl ₂ Solution (50 mM)	10131968	Pass

Assay Name/Specification	Lot # 10140649
<p>PCR Amplification (Hot Start, Human Genomic DNA) A 25 µl reaction in Phusion[®] GC Buffer in the presence of 200 µM dNTPs and 0.5 µM primers containing 50 ng Human Genomic DNA with 0.5 units of Phusion[®] Hot Start Flex DNA Polymerase for 25 cycles of PCR amplification results in the expected 665 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.</p>	Pass
<p>PCR Amplification (7.5 kb Human Genomic DNA) A 50 µl reaction in Phusion[®] HF Buffer in the presence of 200 µM dNTPs and 1.0 µM primers containing 50 ng Human Genomic DNA with 1 unit of Phusion[®] Hot Start Flex DNA Polymerase for 30 cycles of PCR amplification results in the expected 7.5 kb product.</p>	Pass
<p>Endonuclease Activity (Nicking) A 50 µl reaction in NEBuffer 2 in the presence of 200 µM dNTPs containing 1 µg of supercoiled PhiX174 DNA and a minimum of 10 units of Phusion[®] High-Fidelity DNA</p>	Pass

Assay Name/Specification	Lot # 10140649
<p>Polymerase incubated for 4 hours at 37°C and 72°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.</p> <p>PCR Amplification (20 kb Lambda DNA) A 50 µl reaction in Phusion® HF Buffer in the presence of 200 µM dNTPs and 1.0 µM primers containing 10 ng Lambda DNA with 1 unit of Phusion® Hot Start Flex DNA Polymerase for 22 cycles of PCR amplification results in the expected 20 kb product.</p>	<p>Pass</p>

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
08 Feb 2022



Michael Tonello
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08 Feb 2022