

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

Product Name:	Phusion® Hot Start Flex DNA Polymerase
Catalog #:	M0535S/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 74°C.
<i>Lot</i> #:	0061704
Assay Date:	04/2017
Expiration Date:	04/2019
Storage Temp:	-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
Effective Date:	13 Apr 2017

Assay Name/Specification (minimum release criteria)	Lot #0061704
<b>Endonuclease Activity (Nicking)</b> - 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 Polymerase incubated for 4 hours at either 37°C or 72°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>PCR Amplification (20 kb Lambda DNA)</b> - A 50 $\mu$ l reaction in Phusion® HF Buffer in the presence of 200 $\mu$ M dNTPs and 1.0 $\mu$ M 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.	Pass
<b>PCR Amplification (7.5 kb Human Genomic DNA)</b> - A 50 $\mu$ l reaction in Phusion® HF Buffer in the presence of 200 $\mu$ M dNTPs and 1.0 $\mu$ M 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.	Pass
<b>PCR Amplification (Hot Start, Human Genomic DNA)</b> - A 25 $\mu$ l reaction in Phusion® GC Buffer in the presence of 200 $\mu$ M dNTPs and 0.5 $\mu$ M 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.	Pass



M0535S/L Lot: 0061704 Page 1 of 2



240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

\* The BSA in this product has been granted an EDQM "Certificate of Suitability" from the European Directorate for the Quality of Medicines (# R1-CEP-2003-204-Rev00) and has been granted a USDA Certificate for Export of Bovine Blood Plasma/Serum for Manufacture into Pharmaceutical Products.

re

Authorized by Karen Moreira 13 Apr 2017



1 Mm

Inspected by Tony Spear-Alfonso 24 Apr 2017