

New England Biolabs Product Specification

Product Name:	<i>Phusion[®] Hot Start Flex DNA Polymerase</i>
Catalog #:	<i>M0535S/L</i>
Concentration:	<i>2,000 units/ml</i>
Unit Definition:	<i>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>
Shelf Life:	<i>24 months</i>
Storage Temp:	<i>-20°C</i>
Storage Conditions:	<i>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)</i>
Specification Version:	<i>PS-M0535S/L v1.0</i>
Effective Date:	<i>04 Aug 2015</i>

Assay Name/Specification (minimum release criteria)

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 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.

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.

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.

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.

* 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.



Derek Robinson
Director of Quality Control

Date 04 Aug 2015

