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: HindIII-HF® Catalog #: R3104T/M Concentration: 100,000 units/ml

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in 1 hour at 37°C in a total reaction

volume of 50 μ l.

Lot #: 0071711 11/2017 Assay Date: 11/2019 Expiration Date: *-20°C* Storage Temp:

Storage Conditions: 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 500 μg/ml BSA

Specification Version: PS-R3104T/M v1.0

11 Nov 2015 Effective Date:

Assay Name/Specification (minimum release criteria)	Lot #0071711
Endonuclease Activity (Nicking) - A 50 μl reaction in 50 μl PhiX174 NEBuffer 4 60 1 μg HindIII-HF TM PhiX174 37 60 units% conversion to the nicked form as determined by agarose gel electrophoresis. incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Exonuclease Activity (Radioactivity Release) - A 50 μl reaction in CutSmart TM Buffer containing 1 μg of a mixture of single and double-stranded [³ H] <i>E. coli</i> DNA and a minimum of 200 units of HindIII-HF TM incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity) - After a 100-fold over-digestion of Lambda DNA with HindIII-HF TM , >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can be recut with HindIII-HF TM .	Pass
Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart TM Buffer containing 1 µg of Lambda DNA and a minimum of 200 Units of HindIII-HF TM incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE) - HindIII-HF TM is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	Pass

^{*} 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.

Authorized by Derek Robinson

11 Nov 2015

ISO 9001





Inspected by Jianying Luo 20 Dec 2017