

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: Nhel-HF®
Catalog Number: R3131L
Concentration: 20,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg

of Lambda DNA (HindIII digest) in 1 hour at 37°C in a total reaction

volume of 50 μl.

Packaging Lot Number: 10135716
Expiration Date: 01/2024
Storage Temperature: -20°C

Storage Conditions: 250 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50%

Glycerol, 0.15% Triton X-100, 200 µg/ml BSA

Specification Version: PS-R3131S/L v1.0

Nhel-HF® Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
R3131LVIAL	Nhel-HF®	10135715	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10130600	Pass	
B6004SVIAL	rCutSmart™ Buffer	10131975	Pass	

Assay Name/Specification	Lot # 10135716
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda HindIII DNA and a minimum of 200 Units of Nhel-HF™ incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ ³H] E. coli DNA and a minimum of 300 units of Nhel-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 Units of Nhel-HF™ incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Blue-White Screening (Terminal Integrity)	Pass



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Assay Name/Specification	Lot # 10135716
A sample of LITMUS28i vector linearized with a 10-fold excess of Nhel-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	
Ligation and Recutting (Terminal Integrity)  After a 100-fold over-digestion of Lambda HindIII DNA with Nhel-HF™, >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 Nhel-HF™.	Pass

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.

Penghua Zhang **Production Scientist** 

07 Feb 2022

Michael Tonello

Packaging Quality Control Inspector

07 Feb 2022



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