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## New England Biolabs Certificate of Analysis

Product Name: Nhel-HF®
Catalog Number: R3131S
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: 10131695
Expiration Date: 07/2023
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	
R3131SVIAL	Nhel-HF®	10114550	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10126634	Pass	
B6004SVIAL	rCutSmart™ Buffer	10126633	Pass	

Assay Name/Specification	Lot # 10131695
<b>Ligation and Recutting (Terminal Integrity)</b> 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
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) A sample of LITMUS28i vector linearized with a 10-fold excess of Nhel-HF™,	Pass



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Assay Name/Specification	Lot # 10131695
religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	
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

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

17 Dec 2021

Michael Tonello

Packaging Quality Control Inspector

17 Dec 2021



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