NEW ENGLAND BioLabs Inc.
be INSPIRED drive DISCOVERY stay GENUINE
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:	*NheI-HF®*
Catalog Number:	*R3131M*
Concentration:	*100,000 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:	*10122885*
Expiration Date:	*09/2023*
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-R3131M v1.0*

NheI-HF® Component List			
NEB Part Number	**Component Description**	**Lot Number**	**Individual QC Result**
R3131MVIAL	NheI-HF®	10122884	**Pass**
B7024AVIAL	Gel Loading Dye, Purple (6X)	10121392	**Pass**
B6004SVIAL	rCutSmart™ Buffer	10123108	**Pass**

Assay Name/Specification	**Lot # 10122885**
Ligation and Recutting (Terminal Integrity) After a 100-fold over-digestion of Lambda HindIII DNA with NheI-HF™, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16ºC. Of these ligated fragments, >95% can be recut with NheI-HF™.	**Pass**
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 NheI-HF™ incubated for 16 hours at 37ºC 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 [^{3}H] E. coli DNA and a minimum of 300 units of NheI-HF™ incubated for 4 hours at 37ºC releases <0.1% of the total radioactivity.	**Pass**
Blue-White Screening (Terminal Integrity)	**Pass**

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Assay Name/Specification	Lot # 10122885
A sample of LITMUS28i vector linearized with a 10-fold excess of NheI-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	
Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 Units of NheI-HF™ incubated for 4 hours at 37ºC results in <10% conversion to the nicked form 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
18 Oct 2021

Michael Tonello
Packaging Quality Control Inspector
18 Oct 2021