

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: Ncol-HF®
Catalog Number: R3193L
Concentration: 20,000 U/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 μl.

Lot Number: 10045564
Expiration Date: 05/2021
Storage Temperature: -20°C

Storage Conditions: 200 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50%

Glycerol, 200 µg/ml BSA

Specification Version: PS-R3193S/L v1.0

Ncol-HF® Component List			
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result
R3193LVIAL	Ncol-HF®	10045565	Pass
B7204SVIAL	CutSmart® Buffer	10042965	Pass
B7024SVIAL	Gel Loading Dye, Purple (6X)	10038712	Pass

Assay Name/Specification	Lot # 10045564
Blue-White Screening (Terminal Integrity) A sample of LITMUS28i vector linearized with a 10-fold excess of NcoI-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 Units of Ncol-HF™ 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™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 100 units of Ncol-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of Lambda DNA with Ncol-HF™, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated	Pass



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This product has been tested and shown to be in compliance with all specifications.

Production Scientist 26 Apr 2019

detection.

Jay Minichiello

Packaging Quality Control Inspector

23 May 2019



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