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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.

Packaging Lot Number: 10175823
Expiration Date: 11/2024
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				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R3193LVIAL	Ncol-HF®	10172505	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10168649	Pass	
B6004SVIAL	rCutSmart™ Buffer	10173161	Pass	

Assay Name/Specification	Lot # 10175823
Protein Purity Assay (SDS-PAGE) Ncol-HF® is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue	Pass
Blue-White Screening (Terminal Integrity) A sample of LITMUS28i vector linearized with a 10-fold excess of Ncol-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



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Assay Name/Specification	Lot # 10175823
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 fragments, >95% can be recut with Ncol-HF™.	Pass
Non-Specific DNase Activity (16 Hour) A 50 μl reaction in CutSmart™ Buffer containing 1 μg of Lambda DNA and a minimum of 100 Units of Ncol-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.

YunJie Sun \\
Production Scientist

23 Nov 2022

Michael Tonello

Packaging Quality Control Inspector

22 Dec 2022



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