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New England Biolabs Certificate of Analysis

Product Name: NotI-HF®
Catalog Number: R3189S
Concentration: 20,000 U/ml

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

of pBC4 DNA in 1 hour at 37°C in a total reaction volume of 50 μl.

Packaging Lot Number: 10131693
Expiration Date: 07/2023
Storage Temperature: -20°C

Storage Conditions: 50 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-R3189S/L v1.0

NotI-HF® Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R3189SVIAL	NotI-HF®	10116600	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10126634	Pass	
B6004SVIAL	rCutSmart™ Buffer	10130601	Pass	

Assay Name/Specification	Lot # 10131693
Endonuclease Activity (Nicking) A 50 μl reaction in CutSmart™ Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 100 Units of Notl-HF™ incubated for 4 hours at 37°C results in <10%	Pass
conversion to the nicked form as determined by agarose gel electrophoresis. Exonuclease Activity (Radioactivity Release)	Pass
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 200 units of NotI-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	
Ligation and Recutting (Terminal Integrity) After a 10-fold over-digestion of pBC4 DNA with Notl-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 Notl-HF™.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of pBC4 DNA and a minimum of 200 Units of Notl-HF™ incubated for 16 hours at 37°C results in a DNA pattern free	Pass



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Assay Name/Specification	Lot # 10131693
of detectable nuclease degradation as determined by agarose gel electrophoresis	

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.

Penghaa Zhang Production Scientist

14 Dec 2021

Michael Tonello

Packaging Quality Control Inspector

14 Dec 2021



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