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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.

Lot Number: 10054280
Expiration Date: 06/2021
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				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
R3189SVIAL	NotI-HF®	10046577	Pass	
B7204SVIAL	CutSmart® Buffer	10046089	Pass	
B7024SVIAL	Gel Loading Dye, Purple (6X)	10047936	Pass	

Assay Name/Specification	Lot # 10054280
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% 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 200 units of NotI-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> After a 10-fold over-digestion of pBC4 DNA with NotI-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 NotI-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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This product has been tested and shown to be in compliance with all specifications.

Stephanie Cornelio Production Scientist

12 Jun 2019

Jay Minichiello

Packaging Quality Control Inspector

20 Sep 2019