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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: 10069108
Expiration Date: 01/2022
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®	10062598	Pass	
B7204SVIAL	CutSmart® Buffer	10069109	Pass	
B7024SVIAL	Gel Loading Dye, Purple (6X)	10065745	Pass	

Assay Name/Specification	Lot # 10069108
Endonuclease Activity (Nicking) A 50 μl reaction in CutSmart™ Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 100 Units of NotI-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 Notl-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 NotI-HF™ incubated for 16 hours at 37°C results in a DNA pattern free	Pass



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Assay Name/Specification	Lot # 10069108
of detectable nuclease degradation as determined by agarose gel electrophoresis.	

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

Stephanie Cornelio Production Scientist

07 Jan 2020

Jay Minichiello

Packaging Quality Control Inspector

26 Mar 2020

