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New England Biolabs Certificate of Analysis

Product Name:	ApoI-HF
Catalog #:	R3566S/L
Concentration:	20,000 units/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 degrees C in a total reaction volume of 50 μL
<i>Lot</i> #:	0021711
Assay Date:	11/2017
Expiration Date:	11/2019
Storage Temp:	-20°C
Storage Conditions:	200 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 200 μg/ml BSA, (pH 7.4 @ 25°C)
Specification Version:	PS-R3566S/L v1.0
Effective Date:	06 Jan 2016

Assay Name/Specification (minimum release criteria)	Lot #0021711
Blue-White Screening (Terminal Integrity) - A sample of pUC19 vector linearized with a 10-fold excess of ApoI-HF, religated and transformed into an <i>E. coli</i> strain expressing the LacZ beta fragment gene results in $<1\%$ white colonies.	Pass
Exonuclease Activity (Radioactivity Release) - A 50 μ l reaction in CutSmart® Buffer containing 1 μ g of a mixture of single and double-stranded [³ H] <i>E. coli</i> DNA and a minimum of 100 units of ApoI-HF incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Functional Testing (15 minute Digest) - A 50 µl reaction in CutSmart® Buffer containing 1 µg of Lambda DNA and 1 µl of ApoI-HF incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
Ligation and Recutting (Terminal Integrity) - After a 20-fold over-digestion of Lambda DNA with ApoI-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 ApoI-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 ApoI-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
Protein Purity Assay (SDS-PAGE) - ApoI-HF is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass

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Authorized by Derek Robinson 06 Jan 2016



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Inspected by Anthony Francis 03 Nov 2017