

New England Biolabs Certificate of Analysis

Product Name: Apol-HF®
Catalog Number: R3566S
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 rCutSmart Buffer in 1 hour at 37°C in a total reaction volume of 50 µL
Packaging Lot Number: 10260178
Expiration Date: 09/2026
Storage Temperature: -20°C
Storage Conditions: 200 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 200 µg/ml rAlbumin, (pH 7.4 @ 25°C)
Specification Version: PS-R3566S/L v2.0

Apol-HF® Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R3566SVIAL	Apol-HF®	10254302	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10245419	Pass
B6004SVIAL	rCutSmart™ Buffer	10250203	Pass

Assay Name/Specification	Lot # 10260178
Blue-White Screening (Terminal Integrity) A sample of pUC19 vector linearized with a 10-fold excess of Apol-HF®, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 100 units of Apol-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 rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of Apol-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 Apol-HF®, >95% of the DNA	Pass

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fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can be recut with Apol-HF®.	
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of Apol-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) Apol-HF® is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 20 units of Apol-HF® is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	Pass

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

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Nancy Considine
Production Scientist
17 Sep 2024



Talia Monkiewicz
Packaging Quality Control Inspector
19 Sep 2024