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New England Biolabs Certificate of Analysis

Product Name: MluI-HF®
Catalog Number: R3198S
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: 10160251
Expiration Date: 06/2024
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 200 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 200 µg/ml

rAlbumin, 50 % Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-R3198S/L v2.0

Mlul-HF® Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R3198SVIAL	Mlul-HF®	10152651	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10158559	Pass	
B6004SVIAL	rCutSmart™ Buffer	10156429	Pass	

Assay Name/Specification	Lot # 10160251
qPCR DNA Contamination (E. coli Genomic) A minimum of 20 units of Mlul-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
Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of Lambda DNA with Mlul-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 Mlul-HF®.	Pass
Protein Purity Assay (SDS-PAGE) Mlul-HF® is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Non-Specific DNase Activity (16 Hour)	Pass



R3198S / Lot: 10160251

Page 1 of 2

Assay Name/Specification	Lot # 10160251
A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of Mlul-HF® incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	
Functional Test (15 minute Digest) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of Mlul-HF® incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 100 units of Mlul-HF® incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled pUC19 DNA and a minimum of 60 units of Mlul-HF® incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass

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.

Penghua Zhang Production Scientist

10 Jun 2022

Michael Tonello

Packaging Quality Control Inspector

14 Sep 2022



R3198S / Lot: 10160251

Page 2 of 2