

## New England Biolabs Certificate of Analysis

**Product Name:** *MluI-HF*<sup>®</sup>  
**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 1 hour at 37°C in a total reaction volume of 50 µl.  
**Packaging Lot Number:** 10073663  
**Expiration Date:** 09/2021  
**Storage Temperature:** -20°C  
**Storage Conditions:** 200 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-R3198S/L v1.0

MluI-HF <sup>®</sup> Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R3198SVIAL	MluI-HF <sup>®</sup>	10055814	Pass
B7204SVIAL	CutSmart <sup>®</sup> Buffer	10074633	Pass
B7024SVIAL	Gel Loading Dye, Purple (6X)	10059231	Pass

Assay Name/Specification	Lot # 10073663
<b>Protein Purity Assay (SDS-PAGE)</b> MluI-HF is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
<b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of MluI-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
<b>Ligation and Recutting (Terminal Integrity)</b> After a 20-fold over-digestion of Lambda DNA with MluI-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 MluI-HF.	Pass
<b>Functional Test (15 minute Digest)</b> A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of Lambda DNA and 1 µl of MluI-HF incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass

Assay Name/Specification	Lot # 10073663
<p><b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] E. coli DNA and a minimum of 100 units of Mlul-HF incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	<b>Pass</b>
<p><b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in CutSmart™ 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 &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	<b>Pass</b>

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



Penghua Zhang  
Production Scientist  
22 Jun 2020



Michael Tonello  
Packaging Quality Control Inspector  
22 Jun 2020