

New England Biolabs Certificate of Analysis

Product Name: *SacI-HF®*
Catalog Number: *R3156M*
Concentration: *100,000 U/ml*
Unit Definition: *One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA (HindIII digest) in 1 hour at 37°C in a total reaction volume of 50 µl.*
Packaging Lot Number: *10132361*
Expiration Date: *12/2023*
Storage Temperature: *-20°C*
Storage Conditions: *100 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-R3156M v2.0*

SacI-HF® Component List

| NEB Part Number | Component Description | Lot Number | Individual QC Result |
|-----------------|------------------------------|------------|----------------------|
| R3156M VIAL | SacI-HF® | 10132360 | Pass |
| B7024A VIAL | Gel Loading Dye, Purple (6X) | 10127723 | Pass |
| B6004S VIAL | rCutSmart™ Buffer | 10130602 | Pass |

| Assay Name/Specification | Lot # 10132361 |
|--|----------------|
| Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 60 Units of SacI-HF™ incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |
| Blue-White Screening (Terminal Integrity) A sample of LITMUS28i vector linearized with a 10-fold excess of SacI-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 CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 100 units of SacI-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass |
| Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda-HindIII DNA and a | Pass |

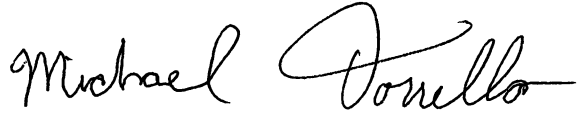
| Assay Name/Specification | Lot # 10132361 |
|---|----------------|
| <p>minimum of 100 Units of SmaI-HF™ incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> | |
| <p>Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of pXba DNA with SmaI-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 SmaI-HF™.</p> | Pass |
| <p>Protein Purity Assay (SDS-PAGE) SmaI-HF™ is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.</p> | 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.



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Production Scientist
18 Jan 2022



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Packaging Quality Control Inspector
18 Jan 2022