

New England Biolabs Certificate of Analysis

Product Name: BbsI-HF[®]
Catalog Number: R3539L
Concentration: 20,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.
Lot Number: 10021240
Expiration Date: 09/2020
Storage Temperature: -20°C
Storage Conditions: 300 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 500 µg/ml BSA, (pH 7.4 @ 25°C)
Specification Version: PS-R3539S/L v1.0

| BbsI-HF [®] Component List | | | |
|-------------------------------------|------------------------------|------------|----------------------|
| NEB Part Number | Component Description | Lot Number | Individual QC Result |
| R3539LVIAL | BbsI-HF [®] | 10020734 | Pass |
| B7204SVIAL | CutSmart [®] Buffer | 10015396 | Pass |
| B7024SVIAL | Gel Loading Dye, Purple (6X) | 10018419 | Pass |

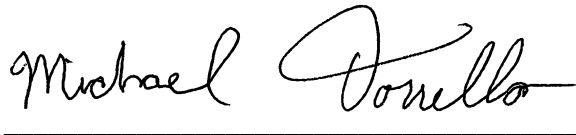
| Assay Name/Specification | Lot # 10021240 |
|--|----------------|
| <p>Exonuclease Activity (Radioactivity Release) A 50 µl reaction in CutSmart[®] Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 100 units of BbsI-HF incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.</p> | Pass |
| <p>Functional Testing (15 minute Digest) A 50 µl reaction in CutSmart[®] Buffer containing 1 µg of Lambda DNA and 1 µl of BbsI-HF incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p> | Pass |
| <p>Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of Lambda DNA with BbsI-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 BbsI-HF.</p> | Pass |
| <p>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 BbsI-HF incubated for 16 hours at 37°C results in a DNA pattern free of</p> | Pass |

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|--|----------------|
| detectable nuclease degradation as determined by agarose gel electrophoresis. | |
| Protein Purity Assay (SDS-PAGE) BbsI-HF is \geq 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| Endonuclease Activity (Nicking) A 50 μ l reaction in CutSmart [®] Buffer containing 1 μ g of supercoiled pUC19 DNA and a minimum of 60 units of BbsI-HF incubated for 4 hours at 37°C results in <20% 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.



Tony Spear-Alfonso
Production Scientist
09 Aug 2018



Michael Tonello
Packaging Quality Control Inspector
21 Sep 2018