

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.
Packaging Lot Number: 10133656
Expiration Date: 09/2023
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 rAlbumin, (pH 7.4 @ 25°C)
Specification Version: PS-R3539S/L v2.0

BbsI-HF [®] Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R3539LVIAL	BbsI-HF [®]	10119528	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10126634	Pass
B6004SVIAL	rCutSmart [™] Buffer	10127377	Pass

Assay Name/Specification	Lot # 10133656
<p>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 BbsI-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>	Pass
<p>qPCR DNA Contamination (E. coli Genomic) A minimum of 20 units of BbsI-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.</p>	Pass
<p>Protein Purity Assay (SDS-PAGE) BbsI-HF[®] is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	Pass
<p>Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of Lambda DNA with BbsI-HF[®], >95% of the DNA</p>	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 BbsI-HF®.	
Functional Testing (15 minute Digest) A 50 µl reaction in rCutSmart™ 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.	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 BbsI-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 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.

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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13 Dec 2021



Michael Tonello
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13 Dec 2021