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New England Biolabs Certificate of Analysis

Product Name: Bbsl-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: 10101565
Expiration Date: 12/2022
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®	10093079	Pass	
B7204SVIAL	CutSmart® Buffer	10097266	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10091035	Pass	

Assay Name/Specification	Lot # 10101565
Exonuclease Activity (Radioactivity Release)	Pass
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.	
Protein Purity Assay (SDS-PAGE)	Pass
BbsI-HF is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	
Non-Specific DNase Activity (16 Hour)	Pass
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 detectable nuclease degradation as determined by agarose gel electrophoresis.	
detectable indicase degradation as determined by agarose ger electrophoresis.	
Endonuclease Activity (Nicking)	Pass
A 50 µl reaction in CutSmart® 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.	



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Assay Name/Specification	Lot # 10101565
Functional Testing (15 minute Digest) A 50 µl reaction in CutSmart® Buffer containing 1 µg of Lambda DNA and 1 µl of Bbsl-HF incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
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,	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**

19 Mar 2021

Michael Tonello

Packaging Quality Control Inspector

19 Mar 2021



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