

New England Biolabs Certificate of Analysis

Product Name: BbsI-HF®
Catalog Number: R3539S
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: 10013145
Expiration Date: 04/2020
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
R3539SVIAL	BbsI-HF®	0031804	Pass
B7204SVIAL	CutSmart® Buffer	3081804	Pass
B7024SVIAL	Gel Loading Dye, Purple (6X)	0251805	Pass

Assay Name/Specification	Lot # 10013145
Endonuclease Activity (Nicking) 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.	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 BbsI-HF incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
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.	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

Assay Name/Specification	Lot # 10013145
<p>>95% can be recut with BbsI-HF.</p> <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 detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	<p>Pass</p>
<p>Protein Purity Assay (SDS-PAGE) BbsI-HF is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<p>Pass</p>

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



Penghua Zhang
Production Scientist
18 Jun 2018



Mary Conlon
Packaging Quality Control Inspector
18 Jun 2018