

## New England Biolabs Certificate of Analysis

**Product Name:** BssSI-v2  
**Catalog Number:** R0680S  
**Concentration:** 10,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in rCutSmart Buffer in 1 hour at 37°C in a total reaction volume of 50 µl.  
**Packaging Lot Number:** 10159547  
**Expiration Date:** 08/2024  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 500 µg/ml rAlbumin, (pH 7.4 @ 25°C)  
**Specification Version:** PS-R0680S/L v3.0

BssSI-v2 Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0680SVIAL	BssSI-v2	10159524	Pass
B6004SVIAL	rCutSmart™ Buffer	10153336	Pass

Assay Name/Specification	Lot # 10159547
<p><b>Exonuclease Activity (Radioactivity Release)</b>            A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] E. coli DNA and a minimum of 100 units of BssSI-v2 incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	Pass
<p><b>Functional Testing (15 minute Digest)</b>            A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of BssSI-v2 incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Non-Specific DNase Activity (16 Hour)</b>            A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 10 units of BssSI-v2 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><b>qPCR DNA Contamination (E. coli Genomic)</b>            A minimum of 10 units of BssSI-v2 is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus.</p>	Pass

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<p>Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is <math>\leq 1</math> E. coli genome.</p>	
<p><b>Protein Purity Assay (SDS-PAGE)</b> BssSI-v2 is <math>\geq 95\%</math> pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>Ligation and Recutting (Terminal Integrity)</b> After a 20-fold over-digestion of Lambda DNA with BssSI-v2, <math>&gt;95\%</math> of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, <math>&gt;95\%</math> can be recut with BssSI-v2.</p>	<b>Pass</b>

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

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