

## New England Biolabs Certificate of Analysis

**Product Name:** *BstEII-HF<sup>™</sup>*  
**Catalog #:** *R3162S/L*  
**Concentration:** *20,000 units/ml*  
**Unit Definition:** *One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in 1 hour at 37°C in a total reaction volume of 50 µl.*  
**Lot #:** *0011509*  
**Assay Date:** *09/2015*  
**Expiration Date:** *9/2017*  
**Storage Temp:** *-20°C*  
**Storage Conditions:** *50 mM KCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA*  
**Specification Version:** *PS-R3162S/L v1.0*  
**Effective Date:** *17 Apr 2013*

Assay Name/Specification (minimum release criteria)	Lot #0011509
<b>Endonuclease Activity (Nicking)</b> - A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 60 units of BstEII-HF <sup>™</sup> incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] <i>E. coli</i> DNA and a minimum of 60 units of BstEII-HF <sup>™</sup> incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	<b>Pass</b>
<b>Ligation and Recutting (Terminal Integrity)</b> - After a 10-fold over-digestion of Lambda DNA with BstEII-HF <sup>™</sup> , >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 BstEII-HF <sup>™</sup> .	<b>Pass</b>
<b>Non-Specific DNase Activity (16 Hour)</b> - A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of Lambda DNA and a minimum of 60 units of BstEII-HF <sup>™</sup> incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	<b>Pass</b>

\* The BSA in this product has been granted an EDQM "Certificate of Suitability" from the European Directorate for the Quality of Medicines (# R1-CEP-2003-204-Rev00) and has been granted a USDA Certificate for Export of Bovine Blood Plasma/Serum for Manufacture into Pharmaceutical Products.

*M. W. Southworth*

*Katrina Francescone*

Authorized by  
Maurice Southworth  
17 Apr 2013

Inspected by  
Katrina Francescone  
06 Oct 2015

