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03 May 2013

Date

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## New England Biolabs Product Specification

Product Name: BstEII-HF®

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  $\mu$ l.

Shelf Life: 24 months Storage Temp:  $-20 \, ^{\circ}\text{C}$ 

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: 03 May 2013

## Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50  $\mu$ l reaction in CutSmart<sup>TM</sup> Buffer containing 1  $\mu$ g of supercoiled PhiX174 DNA and a minimum of 60 units of BstEII-HF<sup>TM</sup> incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in CutSmart<sup>TM</sup> Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup>H] *E. coli* DNA and a minimum of 60 units of BstEII-HF<sup>TM</sup> incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

**Ligation and Recutting (Terminal Integrity)** - After a 10-fold over-digestion of Lambda DNA with BstEII-HF<sup>TM</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>TM</sup>.

Non-Specific DNase Activity (16 Hour) - A 50 μl reaction in CutSmart<sup>TM</sup> Buffer containing 1 μg of Lambda DNA and a minimum of 60 units of BstEII-HF<sup>TM</sup> incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Derek Robinson

Director of Quality Control







<sup>\*</sup> 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.