

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

New England Biolabs Product Specification

| Product Name: | BstBI |
|------------------------|--|
| Catalog #: | R0519S/L |
| Concentration: | 20,000 units/ml |
| Unit Definition: | One unit is defined as the amount of enzyme required to digest 1 μ g of Lambda DNA in 1 hour at 65°C in a total reaction volume of 50 μ l. |
| Shelf Life: | 24 months |
| 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-R0519S/L v1.0 |
| Effective Date: | 20 Aug 2013 |
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Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 μ l reaction in CutSmartTM Buffer containing 1 μ g of supercoiled pUC19 DNA and a minimum of 20 units of BstBI incubated for 4 hours at 65°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 μ l reaction in CutSmartTM Buffer containing 1 μ g of a mixture of single and doublestranded [³H] *E. coli* DNA and a minimum of 100 units of BstBI incubated for 4 hours at 65°C releases <0.1% of the total radioactivity.

Ligation and Recutting (Terminal Integrity) - After a 20-fold over-digestion of Lambda DNA with BstBI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 4 hours at 25°C. Of these ligated fragments, >95% can be recut with BstBI.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart[™] Buffer containing 1 µg of Lambda DNA and a minimum of 20 units of BstBI incubated for 16 hours at 65°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

* 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.

Derek Robinson Director of Quality Control



Date 20 Aug 2013