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New England Biolabs Product Specification

Product Name: BpuEI

Catalog #: R0633S/V
Concentration: 5,000 units/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.

Shelf Life: 24 months
Storage Temp: -20°C

Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 0.32 mM S-adenosylmethionine (SAM), 50% Glycerol,

500 μg/ml rAlbumin (pH 7.4 @ 25°C)

Specification Version: PS-R0633S/V v4.0

Effective Date: 21 Oct 2024

Assay Name/Specification (minimum release criteria)

Ligation and Recutting (Terminal Integrity) - After a 2-fold over-digestion of Lambda DNA with BpuEI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, ~50% can be recut with BpuEI.

Protein Purity Assay (SDS-PAGE) - BpuEI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.

Exonuclease Activity (Radioactivity Release) - A 50 μ l reaction in rCutSmartTM Buffer containing 1 μ g of a mixture of single and double-stranded [3 H] *E. coli* DNA and a minimum of 5 units of BpuEI incubated for 4 hours at 37°C releases <0.5% of the total radioactivity.

Functional Testing (15 minute Digest) - A 50 μl reaction in rCutSmartTM Buffer containing 1 μg of Lambda DNA and 1 μl of BpuEI incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.

Non-Specific DNase Activity (16 Hour) - A 50 μl reaction in rCutSmartTM Buffer containing 1 μg of Lambda DNA and a minimum of 15 units of BpuEI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

qPCR DNA Contamination (*E. coli* Genomic) - A minimum of 5 units of BpuEI is screened for the presence of *E. coli* genomic DNA using SYBR® Green qPCR with primers specific for the *E. coli* 16S rRNA locus. Results are quantified using a standard curve generated from purified *E. coli* genomic DNA. The measured level of *E. coli* genomic DNA contamination is ≤ 1 *E. coli* genome.









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Date 21 Oct 2024

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