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New England Biolabs Product Specification

Product Name:	BccI
Catalog #:	R0704S/L/V
Concentration:	10,000 units/ml
Unit Definition:	One unit is defined as the amount of enzyme required to digest 1 µg of pXba DNA in rCutSmart Buffer supplemented with 2 mM DTT in 1 hour at 37°C in a total reaction volume of 50 µl.
Shelf Life:	24 months
Storage Temp:	-20°C
Storage Conditions:	50mM NaOAc, 500mM NaCl, 0.1mM EDTA, 0.1mM TCEP , 50% Glycerol, 200 ug/ml rAlbumin (pH 6.0 @ 25°C)
Specification Version:	PS-R0704S/L/V v3.0
Effective Date:	19 Aug 2024

Assay Name/Specification (minimum release criteria)

Exonuclease Activity (Radioactivity Release) - A 50 μ l reaction in rCutSmartTM Buffer containing 1 μ g of a mixture of single and double-stranded [³H] *E. coli* DNA and a minimum of 10 units of BccI incubated for 4 hours at 37°C releases <0.3% of the total radioactivity.

Ligation and Recutting (Terminal Integrity) - After a 2-fold over-digestion of pXba DNA with BccI, ~50% 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 BccI.

Non-Specific DNase Activity (16 hour) - A 50 μ l reaction in rCutSmartTM Buffer containing 1 μ g of pXba DNA and a minimum of 10 units of BccI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. NOTE: although no nuclease degradation is detected under these conditions, extended incubations and/or high concentrations of this enzyme may result in star activity. See the product FAQ for recommended reaction conditions for this enzyme.

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

qPCR DNA Contamination (*E. coli* Genomic) - A minimum of 10 units of BccI 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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