

New England Biolabs Product Specification

Product Name:	<i>FspI</i>
Catalog #:	R0135S/L
Concentration:	10,000 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:	10mM Tris-HCl, 300mM NaCl, 0.1mM EDTA, 1mM DTT, 0.15% Triton X-100, 300 ug/ml rAlbumin, 50% Glycerol (pH 7.5 @ 25°C)
Specification Version:	PS-R0135S/L v2.0
Effective Date:	07 Nov 2023

Assay Name/Specification (minimum release criteria)

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA and a minimum of 100 units of FspI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

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

Ligation and Recutting (Terminal Integrity) - After a 10-fold over-digestion of Lambda DNA with FspI, ~50% 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 FspI.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of FspI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Protein Purity Assay (SDS-PAGE) - FspI 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 FspI 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.



New England Biolabs Product Specification

One or more products referenced in this document may be covered by a 3rd-party trademark.
Please visit www.neb.com/trademarks for additional information.



Date 07 Nov 2023

Nancy Considine
Quality Approver

