

New England Biolabs Product Specification

| | |
|-------------------------------|---|
| <i>Product Name:</i> | <i>PshAI</i> |
| <i>Catalog #:</i> | <i>R0593S/L/V</i> |
| <i>Concentration:</i> | <i>10,000 units/ml</i> |
| <i>Unit Definition:</i> | <i>One unit is defined as the amount of enzyme required to digest 1 µg Lambda DNA in rCutSmart™ Buffer in 1 hour at 37°C in a total reaction volume of 50 µl.</i> |
| <i>Shelf Life:</i> | <i>24 months</i> |
| <i>Storage Temp:</i> | <i>-20°C</i> |
| <i>Storage Conditions:</i> | <i>10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml rAlbumin (pH 7.4 @ 25C)</i> |
| <i>Specification Version:</i> | <i>PS-R0593S/L/V v2.0</i> |
| <i>Effective Date:</i> | <i>03 Feb 2022</i> |

Assay Name/Specification (minimum release criteria)

Ligation and Recutting (Terminal Integrity) - After a 10-fold over-digestion of Lambda DNA with PshAI, >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 PshAI.

Endonuclease Activity (Nicking) - A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled pUC19 DNA and a minimum of 30 units of PshAI 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 rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA and a minimum of 100 units of PshAI 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 PshAI 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 rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 50 units of PshAI 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) - PshAI 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 PshAI 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.





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

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 03 Feb 2022

Derek Robinson
Director, Quality Control

