

## New England Biolabs Certificate of Analysis

**Product Name:** PshAI  
**Catalog Number:** R0593S  
**Concentration:** 10,000 U/ml  
**Unit Definition:** 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.  
**Packaging Lot Number:** 10160105  
**Expiration Date:** 08/2024  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml rAlbumin (pH 7.4 @ 25C)  
**Specification Version:** PS-R0593S/L/V v2.0

PshAI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0593SVIAL	PshAI	10160104	Pass
B6004SVIAL	rCutSmart™ Buffer	10156434	Pass

Assay Name/Specification	Lot # 10160105
<b>Non-Specific DNase Activity (16 Hour)</b> 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.	Pass
<b>Functional Testing (15 minute Digest)</b> 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.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] 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.	Pass
<b>Endonuclease Activity (Nicking)</b> 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%	Pass

Assay Name/Specification	Lot # 10160105
conversion to the nicked form as determined by agarose gel electrophoresis.	
<p><b>Protein Purity Assay (SDS-PAGE)</b> PshAI is <math>\geq 95\%</math> pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>qPCR DNA Contamination (E. coli Genomic)</b> 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 <math>\leq 1</math> E. coli genome.</p>	<b>Pass</b>
<p><b>Ligation and Recutting (Terminal Integrity)</b> After a 10-fold over-digestion of Lambda DNA with PshAI, <math>&gt;95\%</math> of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at <math>16^{\circ}\text{C}</math>. Of these ligated fragments, <math>&gt;95\%</math> can be recut with PshAI.</p>	<b>Pass</b>

This product has been tested and shown to be in compliance with all specifications.

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