

## New England Biolabs Certificate of Analysis

**Product Name:** *HpaI*  
**Catalog Number:** *R0105S*  
**Concentration:** *5,000 U/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.*  
**Packaging Lot Number:** *10149795*  
**Expiration Date:** *05/2024*  
**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 @25°C)*  
**Specification Version:** *PS-R0105S/L/V v2.0*

HpaI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0105SVIAL	HpaI	10149794	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10149690	Pass
B6004SVIAL	rCutSmart™ Buffer	10150371	Pass

Assay Name/Specification	Lot # 10149795
<b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 5 units of HpaI 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.	Pass
<b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled pUC19 DNA and a minimum of 15 units of HpaI incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<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 15 units of HpaI 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>Exonuclease Activity (Radioactivity Release)</b>	Pass

Assay Name/Specification	Lot # 10149795
<p>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 50 units of HpaI incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	
<p><b>Protein Purity Assay (SDS-PAGE)</b> HpaI is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>Ligation and Recutting (Terminal Integrity)</b> After a 10-fold over-digestion of Lambda DNA with HpaI, &gt;95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, &gt;95% can be recut with HpaI.</p>	<b>Pass</b>

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

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24 May 2022



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