

## New England Biolabs Certificate of Analysis

**Product Name:** *Hinfl*  
**Catalog Number:** *R0155M*  
**Concentration:** *50,000 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 of 50 µl.*  
**Packaging Lot Number:** *10180768*  
**Expiration Date:** *02/2025*  
**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-R0155T/M v2.0*

Hinfl Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0155MVIAL	Hinfl	10181252	Pass
B6004SVIAL	rCutSmart™ Buffer	10175291	Pass

Assay Name/Specification	Lot # 10180768
<p><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 Hinfl incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	Pass
<p><b>Functional Testing (15 minute Digest)</b>            A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of Hinfl incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Ligation and Recutting (Terminal Integrity)</b>            After a 20-fold over-digestion of Lambda DNA with Hinfl, &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 Hinfl.</p>	Pass
<p><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 100 units of Hinfl incubated for 16 hours at 37°C results in a DNA pattern free of</p>	Pass


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detectable nuclease degradation as determined by agarose gel electrophoresis.	
<p><b>Protein Purity Assay (SDS-PAGE)</b> HinfI is &gt;95% 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 HinfI is screened for the presence of E. coli genomic DNA using SYBR<sup>®</sup> 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>

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

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YunJie Sun  
Production Scientist  
22 Feb 2023



Michael Tonello  
Packaging Quality Control Inspector  
03 Mar 2023