

## New England Biolabs Certificate of Analysis

**Product Name:** HindIII-HF<sup>®</sup>  
**Catalog Number:** R3104M  
**Concentration:** 100,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in 1 hour at 37°C in a total reaction volume of 50 µl.  
**Packaging Lot Number:** 10091706  
**Expiration Date:** 09/2022  
**Storage Temperature:** -20°C  
**Storage Conditions:** 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 500 µg/ml BSA  
**Specification Version:** PS-R3104T/M v1.0

| HindIII-HF <sup>®</sup> Component List |                              |            |                      |
|--|------------------------------|------------|----------------------|
| NEB Part Number                        | Component Description        | Lot Number | Individual QC Result |
| R3104MVIAL                             | HindIII-HF <sup>®</sup>      | 10082129   | Pass                 |
| B7204SVIAL                             | CutSmart <sup>®</sup> Buffer | 10089402   | Pass                 |
| B7024AVIAL                             | Gel Loading Dye, Purple (6X) | 10084973   | Pass                 |

| Assay Name/Specification  | Lot # 10091706 |
|---|----------------|
| <b>Endonuclease Activity (Nicking)</b><br>A 50 µl reaction in 50 µl PhiX174 NEBuffer 4 60 1 µg HindIII-HF <sup>™</sup> PhiX174 37 60 units% conversion to the nicked form as determined by agarose gel electrophoresis. incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass           |
| <b>Exonuclease Activity (Radioactivity Release)</b><br>A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 200 units of HindIII-HF <sup>™</sup> incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.                   | Pass           |
| <b>Ligation and Recutting (Terminal Integrity)</b><br>After a 100-fold over-digestion of Lambda DNA with HindIII-HF <sup>™</sup> , >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 HindIII-HF <sup>™</sup> .  | Pass           |
| <b>Non-Specific DNase Activity (16 Hour)</b><br>A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of Lambda DNA and a minimum of   | Pass           |

| Assay Name/Specification   | Lot # 10091706     |
|--|--------------------|
| <p>200 Units of HindIII-HF™ incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> <p><b>Protein Purity Assay (SDS-PAGE)</b><br/>HindIII-HF™ is &gt;95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.</p> | <p><b>Pass</b></p> |

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

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22 Nov 2020



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