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New England Biolabs Certificate of Analysis

Product Name: HindIII-HF®

Catalog Number: R3104S

Concentration: 20,000 U/mI

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.

Lot Number: 10055523
Expiration Date: 05/2021
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-R3104S/L v1.0

| HindIII-HF® Component List | | | | |
|----------------------------|------------------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| R3104SVIAL | HindIII-HF® | 10044611 | Pass | |
| B7204SVIAL | CutSmart® Buffer | 10046088 | Pass | |
| B7024SVIAL | Gel Loading Dye, Purple (6X) | 10053977 | Pass | |

| Assay Name/Specification | Lot # 10055523 |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------|
| Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 60 Units of HindIII-HF™ incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |
| Protein Purity Assay (SDS-PAGE) HindIII-HF™ is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection. | Pass |
| Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 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. | Pass |
| Ligation and Recutting (Terminal Integrity) After a 100-fold over-digestion of Lambda DNA with HindIII-HF™, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated | Pass |



R3104S / Lot: 10055523

Page 1 of 2

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| fragments, >95% can be recut with HindIII-HF™. | |
| Exonuclease Activity (Radioactivity Release) A 50 μl reaction in CutSmart™ Buffer containing 1 μg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 200 units of HindIII-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass |

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

Jianying Luo Production Scientist

08 May 2019

Jay Minichiello

Packaging Quality Control Inspector

09 Oct 2019

