

## New England Biolabs Certificate of Analysis

**Product Name:** Scal-HF<sup>®</sup>  
**Catalog Number:** R3122L  
**Concentration:** 20,000 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:** 10110093  
**Expiration Date:** 03/2023  
**Storage Temperature:** -20°C  
**Storage Conditions:** 200 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA  
**Specification Version:** PS-R3122S/L v1.0

| Scal-HF <sup>®</sup> Component List |                               |            |                      |
|-------------------------------------|-------------------------------|------------|----------------------|
| NEB Part Number                     | Component Description         | Lot Number | Individual QC Result |
| R3122LVIAL                          | Scal-HF <sup>®</sup>          | 10102479   | Pass                 |
| B7024AVIAL                          | Gel Loading Dye, Purple (6X)  | 10105817   | Pass                 |
| B6004SVIAL                          | rCutSmart <sup>™</sup> Buffer | 10108730   | Pass                 |

| Assay Name/Specification  | Lot # 10110093 |
|---|----------------|
| <p><b>Ligation and Recutting (Terminal Integrity)</b><br/>           After a 2-fold over-digestion of Lambda DNA with Scal-HF<sup>™</sup>, &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 Scal-HF<sup>™</sup>.</p>                                   | Pass           |
| <p><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 60 Units of Scal-HF<sup>™</sup> incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> | Pass           |
| <p><b>Protein Purity Assay (SDS-PAGE)</b><br/>           Scal-HF<sup>™</sup> is &gt;95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.</p>   | Pass           |
| <p><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 100 units of Scal-HF<sup>™</sup> incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>  | Pass           |

| Assay Name/Specification   | Lot # 10110093     |
|--|--------------------|
| <p><b>Endonuclease Activity (Nicking)</b><br/>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 Units of Scal-HF™ incubated for 4 hours at 37°C results in &lt;20% conversion to the nicked form as determined by agarose gel electrophoresis.</p> | <p><b>Pass</b></p> |

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

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