

New England Biolabs Certificate of Analysis

Product Name: Spel-HF[®]
Catalog Number: R3133M
Concentration: 100,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of pXba-XbaI DNA in 1 hour at 37°C in a total reaction volume of 50 µl.
Packaging Lot Number: 10112736
Expiration Date: 06/2023
Storage Temperature: -20°C
Storage Conditions: 250 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 0.15% Triton[®] X-100, 200 µg/ml BSA
Specification Version: PS-R3133M v2.0

| Spel-HF [®] Component List | | | |
|-------------------------------------|-------------------------------|------------|----------------------|
| NEB Part Number | Component Description | Lot Number | Individual QC Result |
| R3133M VIAL | Spel-HF [®] | 10110710 | Pass |
| B7024A VIAL | Gel Loading Dye, Purple (6X) | 10105819 | Pass |
| B6004S VIAL | rCutSmart [™] Buffer | 10111602 | Pass |

| Assay Name/Specification | Lot # 10112736 |
|---|----------------|
| Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart [®] Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 60 units of Spel-HF [®] incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |
| Blue-White Screening (Terminal Integrity) A sample of LITMUS28 vector linearized with a 10-fold excess of Spel-HF [®] , religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies. | Pass |
| Protein Purity Assay (SDS-PAGE) Spel-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 pXba-XbaI digested DNA and a minimum of 100 units of Spel-HF [®] incubated for 16 hours at 37°C results in a DNA | Pass |

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|--|---|
| <p>pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> <p>Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of T7 DNA with Spel-HF[®], >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 Spel-HF[®].</p> <p>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 100 units of Spel-HF[®] incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.</p> | <p style="text-align: center;">Pass</p> <p style="text-align: center;">Pass</p> |

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

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13 Jul 2021



Michael Tonello
Packaging Quality Control Inspector
13 Jul 2021