

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: 10136140
Expiration Date: 11/2023
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 [®]	10126775	Pass
B7024A VIAL	Gel Loading Dye, Purple (6X)	10130600	Pass
B6004S VIAL	rCutSmart [™] Buffer	10132778	Pass

Assay Name/Specification	Lot # 10136140
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 [®] .	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in CutSmart [®] Buffer containing 1 µg of a mixture of single and double-stranded [³ H] 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.	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 pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Endonuclease Activity (Nicking)	Pass

Assay Name/Specification	Lot # 10136140
<p>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.</p>	
<p>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.</p>	Pass
<p>Protein Purity Assay (SDS-PAGE) Spel-HF® is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	Pass

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.



Penghua Zhang
Production Scientist
14 Feb 2022



Josh Hersey
Packaging Quality Control Inspector
14 Feb 2022