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New England Biolabs Certificate of Analysis

Product Name: EcoRI-HF®
Catalog Number: R3101L
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: 10134734
Expiration Date: 11/2023
Storage Temperature: -20°C

Storage Conditions: 300 mM NaCl, 10 mM KPO4, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 0.15

% TritonX-100, 200 μg/ml BSA, (pH 7.0 @ 25°C)

Specification Version: PS-R3101S/L v2.0

EcoRI-HF® Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R3101LVIAL	EcoRI-HF®	10127907	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10121393	Pass	
B6004SVIAL	rCutSmart™ Buffer	10121391	Pass	

Assay Name/Specification	Lot # 10134734
Protein Purity Assay (SDS-PAGE)	Pass
EcoRI-HF™ is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	
Non-Specific DNase Activity (16 Hour)	Pass
A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 Units of EcoRI-HF™ incubated for 16 hours at 37°C results in a DNA pattern free	
of detectable nuclease degradation as determined by agarose gel electrophoresis.	
Ligation and Recutting (Terminal Integrity)	Pass
After a 20-fold over-digestion of Lambda DNA with EcoRI-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 EcoRI-HF™.	
Endonuclease Activity (Nicking)	Pass
A 50 μl reaction in CutSmart [™] Buffer containing 1 μg of supercoiled PhiX174 DNA and	
a minimum of 100 Units of EcoRI-HF™ incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	



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Assay Name/Specification	Lot # 10134734
Blue-White Screening (Terminal Integrity) A sample of pUC19 vector linearized with a 10-fold excess of EcoRI-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
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 EcoRI-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.

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**

10 Jan 2022

Michael Tonello

Packaging Quality Control Inspector

10 Jan 2022



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