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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: 1020303
Expiration Date: 01/2025
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®	10176736	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10193044	Pass	
B6004SVIAL	rCutSmart™ Buffer	10196037	Pass	

Assay Name/Specification	Lot # 10203031
Blue-White Screening (Terminal Integrity) A sample of pUC19 vector linearized with a 10-fold excess of EcoRI-HF™, religated	Pass
and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	
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.	
Exonuclease Activity (Radioactivity Release)	Pass
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.	
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	
Tragments can be ligated with 14 DNA ligase in 10 hours at 10°C. Of these ligated	



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Assay Name/Specification	Lot # 10203031
fragments, >95% can be recut with EcoRI-HF™.	
Non-Specific DNase Activity (16 Hour) 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.	Pass
Protein Purity Assay (SDS-PAGE) EcoRI-HF™ is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	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.

YunJie Sun \
Production Scientist

18 Jan 2023

Michael Tonello

Packaging Quality Control Inspector

11 Aug 2023



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