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New England Biolabs Certificate of Analysis

Product Name: Eagl-HF®
Catalog Number: R3505L
Concentration: 20,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg

of pXba DNA in 1 hour at 37°C in a total reaction volume of 50 μl.

Lot Number:10021292Expiration Date:09/2020Storage Temperature:-20°C

Storage Conditions: 500 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol,

200 μg/ml BSA, (pH 7.4 @ 25°C)

Specification Version: PS-R3505S/L v2.0

Eagl-HF® Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R3505LVIAL	Eagl-HF®	10020266	Pass	
B7204SVIAL	CutSmart® Buffer	10015395	Pass	
B7024SVIAL	Gel Loading Dye, Purple (6X)	10018419	Pass	

Assay Name/Specification	Lot # 10021292
Blue-White Screening (Terminal Integrity) A sample of Litmus38i vector linearized with a 10-fold excess of Eagl-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 Units of Eagl-HF™ incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.	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 100 units of Eagl-HF™ incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of pXba DNA with Eagl-HF™, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments,	Pass



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Assay Name/Specification	Lot # 10021292
>95% can be recut with Eagl-HF™.	
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of pXba DNA and a minimum of 100 Units of Eagl-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) Eagl-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.

Tony Spear-Alfonso Production Scientist

08 Aug 2018

Michael Tonello

Packaging Quality Control Inspector

11 Sep 2018



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