240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Product Specification

Product Name: EcoRI

Catalog #: R0101T/M
Concentration: 100,000 units/ml

Unit Definition:

One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in NEBuffer EcoRI/SspI in 1 hour at

37°C in a total reaction volume of 50 μl.

Shelf Life: 24 months
Storage Temp: -20°C

Storage Conditions: 300 mM NaCl, 10 mM KPO<sub>4</sub>, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 0.15% Triton X-100, 200 µg/ml

*rAlbumin (pH 7.5 @ 25°C)* 

Specification Version: PS-R0101T/M v4.0

Effective Date: 19 Feb 2024

## Assay Name/Specification (minimum release criteria)

**Blue-White Screening (Terminal Integrity)** - A sample of pUC19 vector linearized with a 10-fold excess of EcoRI, religated and transformed into an *E. coli* strain expressing the LacZ beta fragment gene results in <1% white colonies.

Exonuclease Activity (Radioactivity Release) - A 50 μl reaction in 1X NEBuffer EcoRI/SspI containing 1 μg of a mixture of single and double-stranded [ <sup>3</sup>H] *E. coli* DNA and a minimum of 200 units of EcoRI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Ligation and Recutting (Terminal Integrity) - After a 20-fold over-digestion of Lambda DNA with EcoRI, >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.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in 1X NEBuffer EcoRI/SspI containing 1 µg of Lambda DNA and a minimum of 100 units of EcoRI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

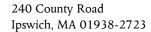
Protein Purity Assay (SDS-PAGE) - EcoRI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.

qPCR DNA Contamination (E. coli Genomic) - A minimum of 20 units of EcoRI is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is  $\leq 1$  E. coli genome.

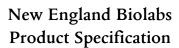








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Date 19 Feb 2024

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