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## New England Biolabs Certificate of Analysis

Product Name: EcoNI
Catalog Number: R0521L
Concentration: 10,000 U/ml

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

of Lambda DNA in rCutSmart Buffer in 1 hour at 37°C in a total

reaction volume of 50 μl.

Packaging Lot Number: 10242443
Expiration Date: 03/2026
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200

μg/ml rAlbumin (pH 7.4 @ 25°C)

Specification Version: PS-R0521S/L v2.0

EcoNI Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
R0521LVIAL	EcoNI	10233805	Pass	
B6004SVIAL	rCutSmart™ Buffer	10235560	Pass	

Assay Name/Specification	Lot # 10242443
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and	Pass
double-stranded [ ³H] E. coli DNA and a minimum of 30 units of EcoNI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	
Functional Testing (15 minute Digest) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of EcoNI incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
Ligation and Recutting (Terminal Integrity) After a 2-fold over-digestion of Lambda DNA with EcoNI, ~50% 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 EcoNI.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 30 units of EcoNI incubated for 16 hours at 37°C results in a DNA pattern free of	Pass



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Assay Name/Specification	Lot # 10242443
detectable nuclease degradation as determined by agarose gel electrophoresis.	
Protein Purity Assay (SDS-PAGE) EcoNI is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic)  A minimum of 10 units of EcoNI 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 ≤ 1 E. coli genome.	Pass

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

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Ana Egana
Production Scientist

06 May 2024

Josh Hersey

Packaging Quality Control Inspector

06 May 2024



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