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New England Biolabs Certificate of Analysis

Product Name: AleI-v2
Catalog Number: R0685L
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: 10242535
Expiration Date: 05/2025
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 200 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 200 µg/ml

rAlbumin, 50 % Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-R0685S/L v2.0

Alel-v2 Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R0685LVIAL	Alel-v2	10241136	Pass	
B6004SVIAL	rCutSmart™ Buffer	10237088	Pass	

Assay Name/Specification	Lot # 10242535
Endonuclease Activity (Nicking) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled pBR322 DNA and	Pass
a minimum of 30 units of AleI-v2 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) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 100 units of Alel-v2 incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity) After a 10-fold over-digestion of Lambda DNA with Alel-v2, >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 Alel-v2.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 50 units of Alel-v2 incubated for 16 hours at 37°C results in a DNA pattern free of	Pass



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Assay Name/Specification	Lot # 10242535	
detectable nuclease degradation as determined by agarose gel electrophoresis.		
Protein Purity Assay (SDS-PAGE) AleI-v2 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 Alel-v2 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.

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.

Ana Egana
Production Scientist

30 May 2024

Josh Hersey

Packaging Quality Control Inspector

30 May 2024



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