

New England Biolabs Certificate of Analysis

Product Name: AvrII
Catalog Number: R0174S
Concentration: 5,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA (HindIII digest) in rCutSmart Buffer in 1 hour at 37°C in a total reaction volume of 50 µl.
Packaging Lot Number: 10131497
Expiration Date: 11/2023
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 500 µg/ml rAlbumin (pH 7.4 @25°C)
Specification Version: PS-R0174S/L/V v2.0

AvrII Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0174SVIAL	AvrII	10128048	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10130600	Pass
B6004SVIAL	rCutSmart™ Buffer	10132770	Pass

Assay Name/Specification	Lot # 10131497
Blue-White Screening (Terminal Integrity) A sample of Litmus28i vector linearized with a 10-fold excess of AvrII, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
Ligation and Recutting (Terminal Integrity) After a 50-fold over-digestion of Lambda HindIII DNA with AvrII, >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 AvrII.	Pass
Protein Purity Assay (SDS-PAGE) AvrII is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 5 units of AvrII 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	Pass

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<p>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.</p>	
<p>Endonuclease Activity (Nicking) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 50 units of AvrII incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p>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 50 units of AvrII incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.</p>	Pass
<p>Functional Testing (15 minute Digest) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of Lambda-HindIII DNA and 1 μl of AvrII incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p>	Pass
<p>Non-Specific DNase Activity (16 Hour) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of Lambda-HindIII DNA and a minimum of 50 units of AvrII incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	Pass

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

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03 Feb 2022



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03 Feb 2022