

be INSPIRED *drive* DISCOVERY *stay* GENUINE

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

New England Biolabs Certificate of Analysis

| Product Name: | AvrII |
|------------------------|---|
| Catalog Number: | R0174L |
| 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: | 10182466 |
| Expiration Date: | 01/2025 |
| 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 | |
| R0174LVIAL | Avrll | 10176960 | Pass | |
| B7024AVIAL | Gel Loading Dye, Purple (6X) | 10182168 | Pass | |
| B6004SVIAL | rCutSmart™ Buffer | 10182169 | Pass | |

| Assay Name/Specification | Lot # 10182466 |
|--|----------------|
| 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 |
| 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. | Pass |
| Exonuclease Activity (Radioactivity Release) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³ H] 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. | Pass |
| Functional Testing (15 minute Digest) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of Lambda-HindIII DNA and 1 μl | Pass |





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| of AvrII incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis. | |
| 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 |
| 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. | 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 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.

YunJie Sun Production Scientist 24 Jan 2023

Michae

Michael Tonello Packaging Quality Control Inspector 02 Jun 2023

