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New England Biolabs Certificate of Analysis

Product Name: AvrII

Catalog Number: R0174S

Concentration: 5,000 U/mI

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

of Lambda DNA (HindIII digest) in 1 hour at 37°C in a total reaction

volume of 50 μl.

Packaging Lot Number: 10100182
Expiration Date: 02/2023
Storage Temperature: -20°C

Storage Conditions: 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50%

Glycerol, 500 μg/ml BSA

Specification Version: PS-R0174S/L v1.0

| AvrII Component List | | | | |
|------------------------|------------------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| R0174SVIAL | AvrII | 10100184 | Pass | |
| B7204SVIAL | CutSmart® Buffer | 10096305 | Pass | |
| B7024AVIAL | Gel Loading Dye, Purple (6X) | 10091035 | Pass | |

| Assay Name/Specification | Lot # 10100182 |
|---|----------------|
| Protein Purity Assay (SDS-PAGE) AvrII is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ 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 |
| Exonuclease Activity (Radioactivity Release) A 50 μl reaction in CutSmart™ 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. | Pass |
| Blue-White Screening (Terminal Integrity) A sample of Litmus28i vector linearized with a 10-fold excess of AvrII, religated | Pass |



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| Assay Name/Specification | Lot # 10100182 |
|--|----------------|
| and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies. | |
| Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ 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 |
| 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 |

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

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Penghua Zhang Production Scientist 09 Mar 2021

Michael Tonello

Packaging Quality Control Inspector

09 Mar 2021

