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New England Biolabs Certificate of Analysis

Product Name: Zral

Catalog Number: R0659L

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 1 hour at 37°C in a total reaction volume of 50 μl.

Lot Number: 10053679
Expiration Date: 09/2021
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-R0659S/L v1.0

| Zral Component List | | | | |
|------------------------|-----------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| R0659LVIAL | Zral | 10053678 | Pass | |
| B7204SVIAL | CutSmart® Buffer | 10053981 | Pass | |

| Assay Name/Specification | Lot # 10053679 |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------|
| Ligation and Recutting (Terminal Integrity) After a 10-fold over-digestion of Lambda DNA with Zral, >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 Zral. | Pass |
| Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 10 Units of Zral 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) Zral is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled M13mp18 DNA and a minimum of 10 units of Zral incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |



R0659L / Lot: 10053679

Page 1 of 2

| Assay Name/Specification | Lot # 10053679 |
|--------------------------------------------------------------------------------------|----------------|
| Exonuclease Activity (Radioactivity Release) | Pass |
| 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 100 units of Zral incubated for 4 | |
| hours at 37°C releases <0.1% of the total radioactivity. | |

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

Jianying Luo Production Scientist

06 Sep 2019

Michael Tonello

Packaging Quality Control Inspector

24 Sep 2019

