

## New England Biolabs Certificate of Analysis

**Product Name:** Ascl  
**Catalog Number:** R0558L  
**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.  
**Packaging Lot Number:** 10072400  
**Expiration Date:** 12/2021  
**Storage Temperature:** -20°C  
**Storage Conditions:** 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA  
**Specification Version:** PS-R0558S/L v1.0

| Ascl Component List |                              |            |                      |
|---------------------|------------------------------|------------|----------------------|
| NEB Part Number     | Component Description        | Lot Number | Individual QC Result |
| R0558LVIAL          | Ascl                         | 10061252   | Pass                 |
| B7204SVIAL          | CutSmart® Buffer             | 10071080   | Pass                 |
| B7024SVIAL          | Gel Loading Dye, Purple (6X) | 10065747   | Pass                 |

| Assay Name/Specification   | Lot # 10072400 |
|--|----------------|
| <b>Blue-White Screening (Terminal Integrity)</b><br>A sample of pNEB193 vector linearized with a 10-fold excess of Ascl, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.  | Pass           |
| <b>Endonuclease Activity (Nicking)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 30 Units of Ascl incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.            | Pass           |
| <b>Exonuclease Activity (Radioactivity Release)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 100 units of Ascl incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass           |
| <b>Ligation and Recutting (Terminal Integrity)</b><br>After a 10-fold over-digestion of Lambda DNA with Ascl, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments,   | Pass           |

| Assay Name/Specification  | Lot # 10072400     |
|---|--------------------|
| <p>&gt;95% can be recut with Ascl.</p> <p><b>Non-Specific DNase Activity (16 Hour)</b><br/>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 30 Units of Ascl incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> | <p><b>Pass</b></p> |

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



Penghua Zhang  
Production Scientist  
20 May 2020



Michael Tonello  
Packaging Quality Control Inspector  
20 May 2020