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: AscI

Catalog #: R0558S/L
Concentration: 10,000 units/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 μ l.

 Lot #:
 0131801

 Assay Date:
 01/2018

 Expiration Date:
 1/2020

 Storage Temp:
 -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
Effective Date: 19 Jul 2017

| Assay Name/Specification (minimum release criteria) | Lot #0131801 |
|---|--------------|
| Blue-White Screening (Terminal Integrity) - A sample of pNEB193 vector linearized with a 10-fold excess of AscI, religated and transformed into an <i>E. coli</i> strain expressing the LacZ beta fragment gene results in <1% white colonies. | Pass |
| Endonuclease Activity (Nicking) - A 50 μl reaction in CutSmart TM Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 30 Units of AscI 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 CutSmart™ Buffer containing 1 μg of a mixture of single and double-stranded [³ H] <i>E. coli</i> DNA and a minimum of 100 units of AscI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass |
| Ligation and Recutting (Terminal Integrity) - After a 10-fold over-digestion of Lambda DNA with AscI, >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 AscI. | Pass |
| Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart TM Buffer containing 1 µg of Lambda DNA and a minimum of 30 Units of AscI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass |

^{*} The BSA in this product has been granted an EDQM "Certificate of Suitability" from the European Directorate for the Quality of Medicines (# R1-CEP-2003-204-Rev00) and has been granted a USDA Certificate for Export of Bovine Blood Plasma/Serum for Manufacture into Pharmaceutical Products.









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Authorized by Derek Robinson 19 Jul 2017







Inspected by Anthony Francis 19 Jan 2018