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: Esp3I
Catalog #: R0734S/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  $\mu$ l.

 Lot #:
 0041803

 Assay Date:
 03/2018

 Expiration Date:
 03/2020

 Storage Temp:
 -20°C

Storage Conditions: 300 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 500 µg/ml BSA, (pH 7.4 @ 25°C)

Specification Version: PS-R0734S/L v1.0 Effective Date: 20 Mar 2018

Assay Name/Specification (minimum release criteria)	Lot #0041803
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 μl reaction in CutSmart® Buffer containing 1 μg of a mixture of single and double-stranded [ <sup>3</sup> H] <i>E. coli</i> DNA and a minimum of 100 units of Esp3I incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
<b>Functional Testing (15 minute Digest)</b> - A 50 μl reaction in CutSmart® Buffer containing 1 μg of Lambda DNA and 1 μl of Esp3I incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> - After a 10-fold over-digestion of Lambda DNA with Esp3I, ≥ 95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, ~75% can be recut with Esp3I.	Pass
Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart® Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of Esp3I incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> - Esp3I is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
RNase Activity (Extended Digestion) - A 10 $\mu$ l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 10 units of Esp3I is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass

Authorized by Derek Robinson 20 Mar 2018







Inspected by Jianying Luo 23 Mar 2018