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## New England Biolabs Certificate of Analysis

Product Name: Smal
Catalog Number: R0141S
Concentration: 20,000 U/ml

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

of Lambda DNA (HindIII digest) in rCutSmart Buffer in 1 hour at 37°C

in a total reaction volume of 50 μl.

Packaging Lot Number: 10162290
Expiration Date: 08/2024
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol,

500 μg/ml rAlbumin (pH 7.4 @ 25°C)

Specification Version: PS-R0141S/L/E v2.0

| Smal Component List    |                              |            |                      |  |
|------------------------|------------------------------|------------|----------------------|--|
| <b>NEB Part Number</b> | <b>Component Description</b> | Lot Number | Individual QC Result |  |
| R0141SVIAL             | Smal                         | 10156748   | Pass                 |  |
| B7024AVIAL             | Gel Loading Dye, Purple (6X) | 10161525   | Pass                 |  |
| B6004SVIAL             | rCutSmart™ Buffer            | 10158558   | Pass                 |  |

| Assay Name/Specification  | Lot # 10162290 |
|---|----------------|
| Functional Testing (15 minute Digest) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of Lambda-HindIII DNA and 1 μl of Smal incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.   | Pass           |
| Protein Purity Assay (SDS-PAGE)<br>Smal is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue<br>detection.   | Pass           |
| Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 60 units of Smal incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass           |
| qPCR DNA Contamination (E. coli Genomic)  A minimum of 20 units of Smal is screened for the presence of E. coli genomic DNA   | Pass           |



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| Assay Name/Specification   | Lot # 10162290 |
|--|----------------|
| using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.                                   |                |
| Exonuclease Activity (Radioactivity Release) A 50 μl reaction in rCutSmart <sup>TM</sup> Buffer containing 1 μg of a mixture of single and double-stranded [ ³H] E. coli DNA and a minimum of 100 units of Smal incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass           |
| Endonuclease Activity (Nicking) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of Smal incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.             | Pass           |
| Blue-White Screening (Terminal Integrity) A sample of pUC19 vector linearized with a 10-fold excess of Smal, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.  | Pass           |
| Ligation and Recutting (Terminal Integrity)  After a 10-fold over-digestion of Lambda DNA with Smal, ~50% 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 Smal.   | Pass           |

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.

Penghaa Zhang Production Scientist

12 Sep 2022

Mary Neal

Packaging Quality Control Inspector

12 Sep 2022



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