

## New England Biolabs Certificate of Analysis

*Product Name:* SmaI  
*Catalog #:* R0141S/L  
*Concentration:* 20,000 units/ml  
*Unit Definition:* One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA (HindIII digest) in 1 hour at 25°C in a total reaction volume of 50 µl.  
*Lot #:* 0841301  
*Assay Date:* 01/2013  
*Expiration Date:* 01/2015  
*Storage Temp:* -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-R0141S/L v1.0  
*Effective Date:* 17 May 2013

| Assay Name/Specification (minimum release criteria)   | Lot #0841301 |
|---|--------------|
| <b>Blue-White Screening (Terminal Integrity)</b> - A sample of pUC19 vector linearized with a 10-fold excess of SmaI, religated and transformed into an <i>E. coli</i> strain expressing the LacZ beta fragment gene results in <1% white colonies.   | <b>Pass</b>  |
| <b>Endonuclease Activity (Nicking)</b> - A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 Units of SmaI incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.                      | <b>Pass</b>  |
| <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 SmaI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.    | <b>Pass</b>  |
| <b>Ligation and Recutting (Terminal Integrity)</b> - After a 10-fold over-digestion of Lambda DNA with SmaI, ~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 SmaI.  | <b>Pass</b>  |
| <b>Non-Specific DNase Activity (16 Hour)</b> - A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 60 Units of SmaI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | <b>Pass</b>  |

\* 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.



Authorized by  
Derek Robinson  
17 May 2013



Inspected by  
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17 May 2013

