

New England Biolabs Product Specification

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| <i>Product Name:</i> | <i>DpnI</i> |
| <i>Catalog #:</i> | <i>R0176S/L</i> |
| <i>Concentration:</i> | <i>20,000 units/ml</i> |
| <i>Unit Definition:</i> | <i>One unit is defined as the amount of enzyme required to digest 1 µg of pBR322 DNA (dam methylated) in 1 hour at 37°C in a total reaction volume of 50 µl.</i> |
| <i>Shelf Life:</i> | <i>24 months</i> |
| <i>Storage Temp:</i> | <i>-20 °C</i> |
| <i>Storage Conditions:</i> | <i>400 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA</i> |
| <i>Specification Version:</i> | <i>PS-R0176S/L v1.0</i> |
| <i>Effective Date:</i> | <i>31 May 2013</i> |

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 20 units of DpnI incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA and a minimum of 200 units of DpnI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Ligation and Recutting (Terminal Integrity) - After a 20-fold over-digestion of pBR322 DNA with DpnI, ~25% 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 DpnI.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart™ Buffer containing 1 µg of pBR322 DNA and a minimum of 100 units of DpnI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Protein Purity Assay (SDS-PAGE) - DpnI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.

* 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.



Derek Robinson
Director of Quality Control

Date 31 May 2013

