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## New England Biolabs Certificate of Analysis

Product Name:	ТspMI
Catalog #:	R0709S/L
Concentration:	5,000 units/ml
Unit Definition:	One unit is defined as the amount of enzyme required to digest 1 μg of pUCAdeno plasmid DNA in 1 hour at 75°C in a total reaction volume of 50 μl.
Lot #:	0031304
Assay Date:	04/2013
Expiration Date:	10/2013
Storage Temp:	-20 °C
Storage Conditions:	300 mM NaCl, 20 mM Tris-HCl (pH 8.0), 1 mM DTT, 1 mM EDTA, 50% Glycerol, 0.10% Triton X-100, 200 μg/ml BSA
Specification Version:	PS-R0709S/L v1.0
Effective Date:	28 Jun 2013

Assay Name/Specification (minimum release criteria)	Lot #0031304
<b>Endonuclease Activity (Nicking)</b> - A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 5 Units of TspMI incubated for 4 hours at 75°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 $\mu$ l reaction in CutSmart <sup>TM</sup> Buffer containing 1 $\mu$ g of a mixture of single and double-stranded [ <sup>3</sup> H] <i>E. coli</i> DNA and a minimum of 50 units of TspMI incubated for 4 hours at 75°C releases <0.1% of the total radioactivity.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> - After a 10-fold over-digestion of pUCAdeno DNA with TspMI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 25°C. Of these ligated fragments, >75% can be recut with TspMI.	Pass
Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart <sup>™</sup> Buffer containing 1 µg of pUCAdeno DNA and a minimum of 5 Units of TspMI incubated for 16 hours at 75°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 28 Jun 2013



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Inspected by David Hough 28 Jun 2013