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: PflMI

Catalog #: R0509S/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 #:
 0291406

 Assay Date:
 06/2014

 Expiration Date:
 6/2016

 Storage Temp:
 -20 °C

Storage Conditions: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA

Specification Version: PS-R0509S/L v1.0
Effective Date: 01 Jul 2014

Assay Name/Specification (minimum release criteria)	Lot #0291406
Endonuclease Activity (Nicking) - A 50 $\mu$ l reaction in NEBuffer 3.1 containing 1 $\mu$ g of supercoiled pNEB193 DNA and a minimum of 50 units of PflMI incubated for 4 hours at 37°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 NEBuffer 3.1 containing 1 $\mu$ g of a mixture of single and double-stranded [ $^3$ H] <i>E. coli</i> DNA and a minimum of 100 units of PflMI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> - After a 10-fold over-digestion of Lambda DNA with PflMI, ~75% 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 PflMI.	Pass
<b>Non-Specific DNase Activity (16 Hour)</b> - A 50 μl reaction in NEBuffer 3.1 containing 1 μg of Lambda DNA and a minimum of 30 Units of PflMI 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> - PflMI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	Pass

<sup>\*</sup> 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.

M.W. Southworth

Authorized by Maurice Southworth 01 Jul 2014







Inspected by Penghua Zhang 01 Jul 2014

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