



BioLabs

1-800-632-7799

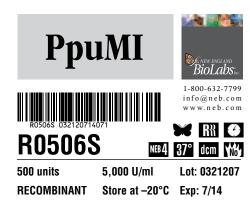
500 units 5.000 U/ml Lot: 0321207 RECOMBINANT Store at -20°C Exp: 7/14

Recognition Site:

5'... R G G W C C Y ... 3' 3′... Y C C W G G R ... 5′

Single Letter Code: R = A or G, W = A or T, Y = C or T

Source: An E. coli strain that carries the cloned PpuMI gene from *Pseudomonas putida* (R. Morgan)



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Single Letter Code: R = A or G, W = A or T, Y = C or T

Source: An E. coli strain that carries the cloned PpuMI gene from *Pseudomonas putida* (R. Morgan)

Supplied in: 50 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme: 10X NFBuffer 4

Reaction Conditions: 1X NEBuffer 4. Incubate at 37°C.

1X NEBuffer 4:

50 mM potassium acetate 20 mM Tris-acetate 10 mM magnesium acetate 1 mM dithiothreitol pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 ug of λ DNA (HindIII digest) in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer B 300 mM NaCl. 10 mM Tris-HCl. 0.1 mM EDTA. 1 mM dithiothreitol, 500 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

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pH 7.9 @ 25°C

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reaction volume of 50 µl.

(pH 7.4). 0.1 mM EDTA. 1 mM dithiothreitol.

Quality Control Assays

Ligation: After 20-fold overdigestion with PpuMI. > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5' termini concentration of $1-2 \mu$ M) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 ul reaction containing 1 µg of DNA and 300 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction incubated for 1 hour with 1 unit of enzyme.

Exonuclease Activity: Incubation of 300 units of enzyme with 1 ug sonicated ³H DNA (10⁵ cpm/ug) for 4 hours at 37°C in 50 µl reaction buffer released < 0.05% radioactivity.

Endonuclease Activity: Incubation of 75 units of enzyme with 1 ug ϕ X174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in 5% conversion to RF II.

Enzyme Properties

Activity in NEBuffers: NEBuffer 1 0%

NEBuffer 2 25% NFBuffer 3 0% NEBuffer 4 100%

When using a buffer other than the optimal (supplied) NEBuffer, it may be necessary to add more enzyme to achieve complete digestion.

Survival in a Reaction: A minimum of 0.13 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: No

Note: Blocked by overlapping *dcm* methylation.

Image: Contract of the second sec

CERTIFICATE OF ANALYSIS

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(HindIII digest) in 1 hour at 37°C in a total