PsiI

1-800-632-7799
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www.neb.com

R0657S 010121014101

R0657S

NEB4 37° Yes

200 units **Lot: 0101210** **Exp: 10/14**
5,000 U/ml **Store at –20°C**

Recognition Site:

5′...T T A▼T A A...3′
3′...A A T▲A T T...5′

Source: An *E. coli* strain that carries the cloned PsiI gene from *Pseudomonas* species SE-G49

More Units

Supplied in: 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 500 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 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 DTT
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of λ DNA 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 DTT, 500 µg/ml BSA and 50% glycerol (pH 7.5 @ 25°C).

Quality Control Assays

Ligation: After 10-fold overdigestion with PsiI, > 95% of DNA fragments can be ligated with T4 DNA Ligase at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of λ DNA and 15 units of PsiI incubated for 16 hours at 37°C resulted in a DNA pattern free of detectable nuclease degradation as determinded by agarose gel electrophoresis.

Exonuclease Activity: Incubation of a 50 µl reaction containing 50 units of PsiI with 1 µg of a mixture of single and double-stranded [^{3}H] *E. coli* DNA (10^5 cpm/µg) for 4 hours at 37°C released < 0.1% of the total radioactivity.

Endonuclease Activity: Incubation of 50 µl reaction containing 15 units of PsiI with 1 µg of pBR322 DNA for 4 hours at 37°C resulted in < 10% conversion to RFII as determined by agarose gel electrophoresis.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	10%
NEBuffer 2	100%
NEBuffer 3	10%
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 0.25 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 65°C for 20 minutes.

Note: Not sensitive to *dam, dcm* or mammalian CpG methylation.

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CERTIFICATE OF ANALYSIS