

PaeR7I



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R0177S 004121014101

R0177S



2,000 units 20,000 U/ml Lot: 0041210

RECOMBINANT Store at -20°C Exp: 10/14

Recognition Site:

5'...CTCGAG...3'
3'...GAGCTC...5'

Source: An *E. coli* strain that carries the cloned PaeR7I gene from *Pseudomonas aeruginosa* PA0303 pMG7 (R. V. Miller)

Supplied in: 50 mM KCl, 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 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 dithiothreitol
pH 7.9 @ 25°C

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

Diluent Compatibility: Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 10-fold overdigestion with PaeR7I, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5' termini concentration of 1–2 µM) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of λ DNA (HindIII Digest) and 60 units of PaeR7I incubated for 16 hours at 37°C resulted in a pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Exonuclease Activity: Incubation of 60 units of PaeR7I with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Endonuclease Activity: Incubation of 50 units of PaeR7I with 1 µg pBR322 plasmid DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 25%
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 0.13 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: No

Notes: The recognition sequence of PaeR7I is identical to that of XhoI. However, it has been determined experimentally that the sequence CTCTCGAG is resistant to cleavage by PaeR7I

Cleavage of mammalian genomic DNA is blocked by CpG methylation.

= Time-Saver™ Qualified (See www.neb.com for details).

CERTIFICATE OF ANALYSIS

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