

PvuII-HF™



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



R3151S 001121014101

R3151S



5,000 units 20,000 U/ml Lot: 0011210
RECOMBINANT Store at -20°C Exp: 10/14

Recognition Site:

5'... CAGCTG... 3'
3'... GTCGAC... 5'

Note: PvuII-HF™ has the same specificity as PvuII (NEB #R0151), but it has been engineered for reduced star activity.

Source: An *E. coli* strain that carries the cloned and modified (T46G) PvuII gene from *Proteus vulgaris* (ATCC 13315)

Supplied in: 200 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 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 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.

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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 7.4 @ 25°C).

Quality Controls

Ligation: After 10-fold overdigestion with PvuII-HF, > 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 DNA and 100 units of enzyme incubated for 16 hours resulted in no degradation of the DNA bands due to nonspecific nucleases.

Exonuclease Activity: Incubation of 500 units of enzyme 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 500 units of enzyme with 1 µg φX174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 10% conversion to RF II.

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Enzyme Properties

Activity in NEBuffers:
NEBuffer 1 0%
NEBuffer 2 25%
NEBuffer 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 1.0 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 80°C for 20 minutes

Plasmid Cleavage: Number of units required to cleave 1 µg of supercoiled plasmid DNA in one hour: pUC19 = 2 units, pBR322 = 4 units.

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

(see other side)

CERTIFICATE OF ANALYSIS

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CERTIFICATE OF ANALYSIS

New icons (see www.neb.com for details)

 = Time-Saver™ Qualified


 = indicates that the enzyme has been engineered


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Page 2 (R3151)

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Page 2 (R3151)