

5,000 units 10,000 U/ml Lot: 0151205 RECOMBINANT Store at -20°C Exp: 5/14

#### **Recognition Site:**

5′... C A G<sup>T</sup>C T G ... 3′ 3′... G T C G A C ... 5′

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

# Also Available In High-Fidelity (HF™) Format



5,000 units 10,000 U/ml Lot: 015120 RECOMBINANT Store at -20°C Exp: 5/14

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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 2.

**Reaction Conditions:** 1X NEBuffer 2. Incubate at 37°C.

## 1X NEBuffer 2:

50 mM NaCl 10 mM Tris-HCl 10 mM MgCl<sub>2</sub> 1 mM DTT pH 7.9 @ 25°C

**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1  $\mu$ g of  $\lambda$  DNA in 1 hour at 37°C in a total reaction volume of 50  $\mu$ l.

## 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 Control Assays

**Ligation:** After 10-fold overdigestion with Pvull, > 95% of the DNA fragments can be ligated with

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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  $\mu$ I reaction containing 1  $\mu$ g of DNA and 10 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  $\mu$ g sonicated <sup>3</sup>H DNA (10<sup>5</sup> cpm/ $\mu$ g) for 4 hours at 37°C in 50  $\mu$ l reaction buffer released < 0.1% radioactivity.

**Endonuclease Activity:** Incubation of 100 units of enzyme with 1  $\mu$ g  $\phi$ X174 RF I DNA for 4 hours at 37°C in 50  $\mu$ I reaction buffer resulted in < 10% conversion to RF II.

# **Enzyme Properties**

Activity in NEBuffers: NEBuffer 1 100% NEBuffer 2 100% NEBuffer 3 100% NEBuffer 4 100%

When using a buffer other than the optimal (supplied) NEBuffer, it may be necessary to add

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

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

#### Heat Inactivation: No

**Plasmid Cleavage:** Number of units required to cleave 1  $\mu$ g of supercoiled plasmid DNA in one hour: pUC19 = 10 units, pBR322 = 10 units.

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

Conditions of low ionic strength, high enzyme concentration, glycerol concentration > 5%, or pH > 8.0 may result in star activity. Pvull does not cut CAGm<sup>4</sup>CTG or CAGm<sup>5</sup>CTG.

### **Companion Products:**

Pvull-HF™	
#R3151S	5,000 Units
#R3151L	25,000 Units
#R3151M	25,000 Units

■ Time-Saver<sup>™</sup> Qualified (See www.neb.com for details).

CERTIFICATE OF ANALYSIS

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