

# Fnu4HI



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R0178S 032121014101

## R0178S



**200 units**      **5,000 U/ml**      **Lot: 0321210**  
**RECOMBINANT**    **Store at -20°C**    **Exp: 10/14**

### Recognition Site:

5'... G C N G C ... 3'  
3'... C G N C G ... 5'

**Source:** An *E. coli* strain that carries the cloned Fnu4HI gene from *Fusobacterium nucleatum* 4H (M. Smith)

Supplied in: 50 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.

**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 20-fold overdigestion with Fnu4HI, < 5% 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 50 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 approximately 10 units of enzyme with 1 µg sonicated <sup>3</sup>H DNA (10<sup>5</sup> cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

### Enzyme Properties

**Activity in NEBuffers:**  
NEBuffer 1    10%  
NEBuffer 2    25%  
NEBuffer 3    25%  
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:** 30 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

**Notes:** Fnu4HI produces DNA fragments that have a single-base 5' extension which are more difficult to ligate than blunt-ended fragments.

Cleavage of mammalian genomic DNA is blocked by overlapping CpG methylation.

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

CERTIFICATE OF ANALYSIS

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