

NdeI



1-800-632-7799
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R0111S 029120614061



R0111S

4,000 units 20,000 U/ml Lot: 0291206
RECOMBINANT Store at -20°C Exp: 6/14

Recognition Site:

5'... C A T A T G ... 3'
3'... G T A T A C ... 5'

Source: An *E. coli* strain that carries the cloned NdeI gene from *Neisseria denitrificans* (NRCC 31009)

Supplied in: 100 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)

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Quality Control Assays

Ligation: After 20-fold overdigestion with NdeI, > 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 75 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 4,000 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 30 units of enzyme with 1 µg φX174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 15% conversion to RF II.

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Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 75%
NEBuffer 2 100%
NEBuffer 3 75%
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: 50 units of enzyme were inactivated by incubation at 65°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 = 3 units.

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

DNA purified by standard miniprep procedures is cleaved at lower rates.

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

CERTIFICATE OF ANALYSIS

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