NaeI







R0190S



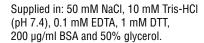
500 units 10,000 U/ml Lot: 0581212 RECOMBINANT Store at -20°C Exp: 12/14

Recognition Site:

5′...GCC GGC...3′ 3'... C G G C C G ... 5'

Source: An E. coli strain that carries the cloned Nael gene from *Nocardia aerocolonigenes* (ATCC 23870)

New Reaction Buffer



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 pXba DNA in 1 hour at 37°C in a total reaction volume of 50 ul.

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 Nael. approximately 75% 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 ul reaction containing 1 µg of DNA and 100 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 50 units of enzyme with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 ul reaction buffer released < 1.0% radioactivity.

Endonuclease Activity: Incubation of 50 units of enzyme with 1 ug ϕ X174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 5% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 75% NEBuffer 2 NFBuffer 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 0.50 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 40 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: Demonstrates marked site preferences and cuts pBR322 DNA very slowly. NgoMIV. an isoschizomer of Nael, exhibits less site preference.

Cleavage of mammalian genomic DNA is blocked by CpG methylation.

U.S. Patent No. 5,292,651

CERTIFICATE OF ANALYSIS

NaeI



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