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R0588S 013120813081






R0588S

100 units 2,000 U/ml Lot: 0131208

RECOMBINANT Store at -70°C Exp: 8/13

Recognition Site:

5'... GGCCGGCC... 3'

3'... CCGGCCGG... 5'

Source: An *E. coli* strain that carries the cloned FseI gene from *Frankia* species Eul1b (NRRL 18528)

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, 100X BSA.

Reaction Conditions: 1X NEBuffer 4, supplemented with 100 µg/ml BSA. 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 Adenovirus-2 DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

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 FseI, > 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 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 50 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 50 units of enzyme with 1 µg φX174 RF I DNA for 4 hours at 37°C in a 50 µl reaction buffer resulted in < 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:


NEBuffer 1	100%
NEBuffer 2	75%
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 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: Impaired by some combinations of overlapping *dcm* methylation. Cleavage of mammalian genomic DNA is blocked by CpG methylation.

 = Time-Saver™ Qualified (See www.neb.com for details).
U.S. Patent No. 5,543,308

CERTIFICATE OF ANALYSIS




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