BsmFI

1-800-632-7799
info@neb.com
www.neb.com

R0572S 024120614061

R0572S

100 units **2,000 U/ml** **Lot: 0241206**

RECOMBINANT **Store at –20°C** **Exp: 6/14**

Recognition Site:

5′. . . G G G A C (N)$_{10}$▼. . . 3′
3′. . . C C C T G (N)$_{14}$▲. . . 5′

Source: An *E. coli* strain that carries the cloned BsmFI gene from *Bacillus stearothermophilus* F (ER2683)

Supplied in: 50 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

Now Recombinant

Reagents Supplied with Enzyme:
10X NEBuffer 4, 100X BSA.

Reaction Conditions: 1X NEBuffer 4 supplemented with 100 µg/ml BSA.
Incubate at 65°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 of pBR322 DNA in 1 hour at 65°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 5-fold overdigestion with BsmFI, > 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 pBR322 DNA and 5 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 100 units of enzyme with 1 µg sonicated ^{3}H DNA (10^5 cpm/µg) for 4 hours at 65°C in 50 µl reaction buffer released < 0.1% radioactivity.

Endonuclease Activity: Incubation of 10 units of enzyme with 1 µg pUC19 plasmid DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers

NEBuffer 1	25%
NEBuffer 2	50%
NEBuffer 3	50%
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: Suitable for an extended or overnight digestion. Enzyme is active > 8 hours.

Heat Inactivation: 80°C for 20 minutes.

Plasmid Cleavage: Number of units required to cleave 1 µg of supercoiled plasmid DNA in one hour: 2 units.

Notes: BsmFI is an isoschizomer of FinI. Occasionally, BsmFI has been shown to cleave the sequence GGGAC(9/13). The exact frequency of this occurrence has yet to be determined.

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

Incubation at 37° results in 50% activity.

Incubation longer than 4 hours is not recommended.

Companion Products:

dam$^-$/dcm$^-$ Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

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

CERTIFICATE OF ANALYSIS

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