BsoBI

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R0586S 015121014101

R0586S

10,000 units **10,000 U/ml** **Lot: 0151210**
RECOMBINANT **Store at –20°C** **Exp: 10/14**

Recognition Site:

5′...C▼Y C G R G...3′
3′...G R G C Y▲C...5′

Single Letter Code: R = A or G, Y = C or T

Source: An *E. coli* strain that carries the cloned BsoBI gene from *Bacillus stearothermophilus* JN2091 (D. Clark)

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

Reagents Supplied with Enzyme:
10X NEBuffer 2.

Reaction Conditions: 1X NEBuffer 2. Incubate at 37°C.

1X NEBuffer 2:
50 mM NaCl
10 mM Tris-HCl
10 mM $MgCl_2$
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 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 dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 50-fold overdigestion with BsoBI, > 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 λ DNA and 40 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 160 units of enzyme with 1 µg sonicated ^3H DNA (10^5 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	**100%**
NEBuffer 3	100%
NEBuffer 4	50%

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: No

Notes: BsoBI is a thermophilic isoschizomer of AvaI.

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

The recommended incubation temperature has been changed from 65°C to 37°C to minimize star activity.

Conditions of low ionic strength, high enzyme concentration, glycerol concentration > 5% or pH > 8.0 may result in star activity.

U. S. Patent NO. 5,492,823

CERTIFICATE OF ANALYSIS

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