Bsp1286I

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

R0120S 0191207 14071

R0120S

NEB4 BSA 37° Yes

500 units **Lot: 0191207** **Exp: 7/14**
5,000 U/ml **Store at –20°C**

Recognition Site:

5′... G D G C H▼C ...3′
3′... C▲H C G D G ...5′

Single Letter Code: D = A or G or T,
H = A or C or T

Source: An *E. coli* strain that carries the cloned Bsp1286I gene from *Bacillus sphaericus* (IAM 1286)

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 400 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 λ DNA in 1 hour at 37°C in 50 µl of reaction buffer.

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). Supplement with additional 200 µg/ml BSA when making dilutions with Diluent Buffer A.

Quality Control Assays

Ligation: After 10-fold overdigestion with Bsp1286I, > 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 in NEBuffer 4 containing 1 µg of λ DNA and a minimum of 5 units of enzyme incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Exonuclease Activity: Incubation of 50 units of enzyme with 1 µg sonicated ^{3}H 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	25%
NEBuffer 2	25%
NEBuffer 3	25%
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: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 35 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: Bsp1286I recognizes six distinct sequences, GGGCCC, GAGCCC, (complement GGGCTC), GTGCCC, (complement GGGCAC), GAGCAC, GTGCAC, and GAGCTC (complement GTGCTC).

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

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

nqa ISO 9001 Registered Quality Management
nqa ISO 14001 Registered Environmental Management
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CERTIFICATE OF ANALYSIS

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