BstNI

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

R0168S 015121014101

R0168S

NEB2 BSA 60° No

3,000 units	**Lot: 0151210**	**Exp: 10/14**
RECOMBINANT	**10,000 U/ml**	**Store at –20°C**

Recognition Site:

5′... C C▼W G G ...3′
3′... G G W▲C C ...5′

Single Letter Code: W = A or T

Source: An *E. coli* strain that carries the cloned BstNI gene from *Bacillus stearothermophilus* N (D. Comb)

More Units, Now Recombinant

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, 100X BSA.

Reaction Conditions: 1X NEBuffer 2, supplemented with 100 µg/ml BSA.
Incubate at 60°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 60°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 2-fold overdigestion with BstNI, < 5% of the DNA fragments can be ligated.

16-Hour Incubation: A 50 µl reaction containing 1 µg of 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 100 units of enzyme with 1 µg sonicated ^3H DNA (10^5 cpm/µg) for 4 hours at 60°C in 50 µl reaction buffer released < 0.2% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	10%
NEBuffer 2	**100%**
NEBuffer 3	100%
NEBuffer 4	75%

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 1.00 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: No

Notes: BstNI is an isoschizomer of EcoRII but cuts DNA at a different location. (EcoRII cuts before the two C residues).

BstNI produces DNA fragments that have a single-base 5′ extension which are more difficult to ligate than blunt-ended fragments.

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

Incubation at 37°C results in 30% activity.

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

CERTIFICATE OF ANALYSIS

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