





R0113S



Recognition Site:

5′... C C A N N N N N T G G ... 3′ 3′... G G T N N N N N A C C ... 5′

Source: An *E. coli* strain that carries the cloned BstXI gene from *Bacillus stearothermophilus* X1 (N. Welker)

Now Recombinant

Supplied in: 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 500 μg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme: 10X NFBuffer 3.

Reaction Conditions: 1X NEBuffer 3. Incubate at 37°C.

1X NEBuffer 3: 100 mM NaCl 50 mM Tris-HCl 10 mM MgCl₂ 1 mM DTT pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 μ g of λ DNA in 1 hour at 37°C in a total reaction volume of 50 μ l.

Diluent Compatibility: Diluent Buffer B 300 mM NaCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 500 μg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 20-fold overdigestion with BstXI, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5´ termini concentration of 1–2 μ M) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 μ I reaction containing 1 μ g 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 100 units of enzyme with 1 μ g sonicated ³H DNA (2 x10⁵ cpm/ μ g) for 4 hours at 37°C in 50 μ l reaction buffer released < 0.2% radioactivity.

Endonuclease Activity: Incubation of 100 units of enzyme with 1 μ g pUC19 plasmid DNA for 4 hours at 37°C in 50 μ l reaction buffer resulted in < 5% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 0% NEBuffer 2 50% NEBuffer 3 **100%** NEBuffer 4 20%

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

Heat Inactivation: 65°C for 20 minutes.

Notes: Blocked by some combinations of overlapping *dcm* methylation.

Companion Products:

dam⁻/dcm⁻ Competent E. coli

#C2925H 20 transformation reactions #C2925| 24 transformation reactions

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

CERTIFICATE OF ANALYSIS

BstXI



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

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