

Lot: 0031207 500 units 5.000 U/ml RECOMBINANT Store at -20°C Exp: 7/14

Recognition Site:

5′...GTCTCN^NN...3′ 3′... CAGAGNN....5′

Description: Nt.BsmAl is a nicking endonuclease that cleaves only one strand of DNA on a doublestranded DNA substrate.

Source: An *E. coli* strain expressing an altered form of the BsmAI restriction genes from Bacillus stearothermophilus A664





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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.

Reagents Supplied with Enzyme: 10X NFBuffer 4

Reaction Conditions: 1X NEBuffer 4. Incubate at 37°C.

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1 mM DTT

pH 7.9 @ 25°C

20 mM Tris acetate

50 mM potassium acetate

10 mM magnesium acetate

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Unit Definition: One unit is defined as the amount of enzyme required to convert 1 ug of supercoiled plasmid DNA to open circular form 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).

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Diluent Compatibility: Diluent Buffer A

1 mM dithiothreitol, 200 µg/ml BSA and

50% glycerol (pH 7.4 @ 25°C).

50 mM KCI. 10 mM Tris-HCI. 0.1 mM EDTA.

Quality Control Assays

16-Hour Incubation: A 50 ul reaction containing 1 ug of DNA and 40 units of enzyme incubated for 16 hours showed no degradation of DNA fragments.

Exonuclease Activity: Incubation of 40 units of enzyme with 1 µg sonicated [3H] DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.05% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 100% NEBuffer 2 50% NEBuffer 3 10% 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: 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.

Companion Products:

Nb.BbvCl #R0631S #R0631L	1,000 units 5,000 units
Nt.Alwl #R0627S #R0627L	500 units 2,500 units
Nt.BstNBI #R0607S #R0607L	1,000 units 5,000 units

References:

- 1. Song, Q. et al. (2010). Anal. Chem. [Epub ahead of print].
- 2. Zhang, P. et al. (2010) Protein Expr. Purif. 69, 226-234. [Epub 2009 Sep 9].

CERTIFICATE OF ANALYSIS

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Nt.BstNBI #R0607S #R0607L	1,000 units 5,000 units

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0.1 mM EDTA. 1 mM dithiothreitol. 200 ug/ml BSA and 50% glycerol. BioLabs **Reagents Supplied with Enzyme:** 10X NFBuffer 4 1-800-632-7799 info@neb.com www.neb.com Reaction Conditions: 1X NFBuffer 4.