Nt.BbvCI







R0632S

RX NEB 4 37° Yes

1.000 units 10.000 U/ml Lot: 0031211 RECOMBINANT Store at -20°C Exp: 11/14

Recognition Site:

5'... C CT C A G C ... 3' 3'... GGAGTCG...5'

Description: Nt.BbvCl 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 BbvCl restriction genes [Ra(K169E):Rb+] from Bacillus brevis (L. Ge)

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 NFBuffer 4.

Reaction Conditions: 1X NEBuffer 4. 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 convert 1 ug of supercoiled plasmid DNA to open circular form in 1 hour at 37°C in a total reaction volume of 50 ul.

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

16-Hour Incubation: A 50 ul reaction containing 1 µg of DNA and 30 units of enzyme incubated for 16 hours showed no degradation of DNA fragments.

Exonuclease Activity: Incubation of 30 units of enzyme with 1 µg sonicated [3H] DNA (105 cpm/µg) for 4 hours at 37°C in 50 ul reaction buffer released < 0.05% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 50% NEBuffer 2 100% 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: 50 units of enzyme were inactivated by incubation at 80°C for 20 minutes.

Note: The nomenclature of this enzyme has been changed.

Companion Products:

Nb.BbvCI (NEB #R0631)

5'... CCTCAGC...3' 3'... GGAGT,CG...5'

Nt.BstNBI (NEB #R0607)

3'... CTCAGNNNNN ...5'

Nt.AlwI (NEB #R0627)

5'... GGATCNNNNN ... 3' 3'... CCTAGNNNNN ... 5'

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

Nt.BbvCI



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

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5′ GAGTCNNNN ...3′ 3'... CTCAGNNNNN ...5'

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