Nb.BsrDI





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

R0648S ➤ RN NE2 65° West







1,000 units 10.000 U/ml Lot: 0041210 RECOMBINANT Store at -20°C Exp: 10/14

Recognition Site:

5'... G C A A T G N N ... 3' 3'... C G T T A C N N ... 5'

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

Source: An *E. coli* strain expressing only the large subunit of the BsrDI restriction gene from *Bacillus* stearothermophilus D70 (Z.Chen).

Supplied in: 150 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM dithiothreitol and 50% alvcerol.

Reagents Supplied with Enzyme:

10X NFBuffer 2

Reaction Conditions: 1X NEBuffer 2. Incubate at 65°C.

1X NEBuffer 2:

10 mM Tris-HCI 10 mM MgCl_o 50 mM NaCl 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 pUC19 DNA to open circular form in 1 hour at 65°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 40 units of enzyme incubated for 16 hours showed no degradation of DNA fragments.

Exonuclease Activity: Incubation of 100 units of enzyme with 1 µg sonicated [3H] DNA (205 cpm/µg) for 4 hours at 65°C in 50 ul reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 10% NEBuffer 2 100% NEBuffer 3 25% NEBuffer 4 50%

When using a buffer other than the optimal (supplied) NEBuffer, it may be necessary to add more enzyme to achieve complete nicking.

Survival in a Reaction: A minimum of 0.25 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 100 units of enzyme were inactivated by incubation at 80°C for 20 minutes.

Note: Incubation at 37°C results in 20% activity. Not sensitive to dam, dcm or mammalian CpG methylation.

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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R0648S ➤ R3 E2 E5 Mg











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CERTIFICATE OF ANALYSIS