

R0570S NEB 4 BSA 65° 🐝

100

1.000 units Lot: 0051208 Exp: 8/14 10.000 U/ml Store at -20°C

Recognition Site:

5′... G W G C W C ... 3′ 3′... C_•W C G W G ... 5′

Single Letter Code: W = A or T

Source: Bacillus stearothermophilus (P. Shaw)

New Reaction Buffer



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Source: Bacillus stearothermophilus (P. Shaw)

New Reaction Buffer

Supplied in: 100 mM NaCl, 50 mM Tris-HCl (pH 7.6), 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme: 10X NEBuffer 4, 100X BSA.

Reaction Conditions: 1X NEBuffer 4, supplemented with 100 µg/ml BSA. Incubate at 65°C.

1X NEBuffer 4:

50 mM potassium acetate 20 mM Tris-acetate 10 mM magnesium acetate 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 65°C in a total reaction volume of 50 µl.

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

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Quality Control Assays

Ligation: After 10-fold overdigestion with BsiHKAI, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5' termini concentration of $1-2 \mu M$) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 100 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 $(10^5 \text{ cpm/}\mu\text{g})$ for 4 hours at 65°C in 50 μI reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers: NEBuffer 1 50% NEBuffer 2 100%

NEBuffer 3 100% 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: Not recommended for digest over 1 hour.

Heat Inactivation: No

Note: Bsi HKAI is an isoschizomer of HgiAI.

Not sensitive to dam, dcm or mammalian CpG methylation.

Incubation at 37°C results in 5% activity.

CERTIFICATE OF ANALYSIS

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