BsaBI

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

R0537S 010120914091

R0537S

2,000 units **Lot: 0101209** **Exp: 9/14**
10,000 U/ml **Store at –20°C**

Recognition Site:

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

Source: *Bacillus stearothermophilus* B674 (Z. Chen)

New Reaction Buffer
New Storage Conditions

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

Reagents Supplied with Enzyme:
10X NEBuffer 4.

Reaction Conditions: 1X NEBuffer 4.
Incubate at 60°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 (*dam*⁻) in 1 hour at 60°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 dithiothreitol, 500 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 10-fold overdigestion with BsaBI, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5′ termini concentration of 1–2 µM) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 50 units of enzyme incubated for 16 hours resulted in no degradation of the DNA bands due to nonspecific nucleases. However, fragments produced by noncanonical cleavage due to star activity may be observed with 10 units of enzyme in similar conditions.

Exonuclease Activity: Incubation of 100 units of enzyme with 1 µg sonicated ^{3}H DNA (10^{5} cpm/µg) for 4 hours at 60°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
NEBuffer 2	**100%**
NEBuffer 3	75%
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: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 80°C for 20 minutes.

Notes: Blocked by overlapping *dam* methylation.

Cleavage of mammalian genomic DNA is blocked by some combinations of overlapping CpG methylation.

Incubation at 37°C results in 20% activity.

Conditions of high enzyme concentration, glycerol concentration > 5% or pH > 8.0 may result in star activity.

Companion Products:
dam⁻/*dcm*⁻ Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

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

CERTIFICATE OF ANALYSIS

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