BmgBI

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

R0628S 006120814081

R0628S

NEB3 BSA 37° Yes

500 units **10,000 U/ml** **Lot: 0061208**

RECOMBINANT **Store at –20°C** **Exp: 8/14**

Recognition Site:

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

Source: An *E. coli* strain that carries the BmgBI gene from *Bacillus megaterium* (T. Le)

Supplied in: 200 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

New Diluent Buffer

Reagents Supplied with Enzyme:
10X NEBuffer 3, 100X BSA.

Reaction Conditions: 1X NEBuffer 3, supplemented with 100 µg/ml BSA. Incubate at 37°C.

1X NEBuffer 3:
100 mM NaCl
50 mM Tris-HCl
10 mM $MgCl_2$
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 37°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 2-fold overdigestion with BmgBI, approximately 75% of the DNA fragments can be ligated. Of these, approximately 50% can be recut with BmgBI due to the non-palindromic recognition site. The remaining products are recleavable by AatII or PmlI.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 50 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 50 units of enzyme with 1 µg sonicated [^{3}H] DNA (10^5 cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	0%
NEBuffer 2	25%
NEBuffer 3	**100%**
NEBuffer 4	10%

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: 20 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Note: BmgBI is an isoschizomer of BtrI.

When DNA is cut with BmgBI and then ligated, only 50% of these ligated sites regenerate BmgBI sites because its recognition site is non-palindromic.

Cleavage of mammalian genomic DNA is blocked by CpG methylation.

(see other side)

CERTIFICATE OF ANALYSIS

BmgBI

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

R0628S 006120814081

R0628S

NEB3 BSA 37° Yes

500 units 10,000 U/ml Lot: 0061208

RECOMBINANT Store at –20°C Exp: 8/14

Recognition Site:

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

Source: An *E. coli* strain that carries the BmgBI gene from *Bacillus megaterium* (T. Le)

Supplied in: 200 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

New Diluent Buffer

Reagents Supplied with Enzyme:
10X NEBuffer 3, 100X BSA.

Reaction Conditions: 1X NEBuffer 3, supplemented with 100 µg/ml BSA. Incubate at 37°C.

1X NEBuffer 3:
100 mM NaCl
50 mM Tris-HCl
10 mM $MgCl_2$
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 37°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 2-fold overdigestion with BmgBI, approximately 75% of the DNA fragments can be ligated. Of these, approximately 50% can be recut with BmgBI due to the non-palindromic recognition site. The remaining products are recleavable by AatII or PmlI.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 50 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 50 units of enzyme with 1 µg sonicated [^{3}H] DNA (10^5 cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	0%
NEBuffer 2	25%
NEBuffer 3	**100%**
NEBuffer 4	10%

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: 20 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Note: BmgBI is an isoschizomer of BtrI.

When DNA is cut with BmgBI and then ligated, only 50% of these ligated sites regenerate BmgBI sites because its recognition site is non-palindromic.

Cleavage of mammalian genomic DNA is blocked by CpG methylation.

(see other side)

CERTIFICATE OF ANALYSIS

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

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

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

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