# BsrBI

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

R0102S 006120814081

## R0102S

**1,000 units** **Lot: 0061208** **Exp: 8/14**
**10,000 U/ml** **Store at –20°C**

**Recognition Site:**

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

**Source:** *Bacillus stearothermophilus* CPW193 (Z. Chen)

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

**New Reaction Buffer**

**Reagents Supplied with Enzyme:**
10X NEBuffer 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 digest 1 µg of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

**Diluent Compatibility:** Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

## Quality Control Assays

**Ligation:** After 10-fold overdigestion with BsrBI, approximately 75% 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, approximately 50% can be recut.

**16-Hour Incubation:** A 50 µl reaction containing 1 µg of DNA and 60 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 80 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 | 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:** A minimum of 0.5 unit is required to digest 1 µg of substrate DNA in 16 hours.

**Heat Inactivation:** 80°C for 20 minutes.

**Notes:** When DNA is cut with BsrBI and then ligated, only 50% of these ligated sites regenerate BsrBI sites because its recognition sequence is non-palindromic. Ligated sites not recleavable by BsrBI are recleavable by either SacI or SacII. BsrBI is fully active at 50°C.

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

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

CERTIFICATE OF ANALYSIS

BsrBI

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

R0102S 006120814081

R0102S

1,000 units Lot: 0061208 Exp: 8/14
10,000 U/ml Store at –20°C

Recognition Site:

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

Source: *Bacillus stearothermophilus* CPW193 (Z. Chen)

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

New Reaction Buffer

Reagents Supplied with Enzyme:
10X NEBuffer 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 digest 1 µg of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 10-fold overdigestion with BsrBI, approximately 75% 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, approximately 50% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 60 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 80 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 | 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: A minimum of 0.5 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 80°C for 20 minutes.

Notes: When DNA is cut with BsrBI and then ligated, only 50% of these ligated sites regenerate BsrBI sites because its recognition sequence is non-palindromic. Ligated sites not recleavable by BsrBI are recleavable by either SacI or SacII. BsrBI is fully active at 50°C.

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

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

CERTIFICATE OF ANALYSIS