BciVI

NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com

R0596S 014121114111

R0596S

NEB4 37° Yes

200 units **10,000 U/ml** **Lot: 0141211**

RECOMBINANT **Store at –20°C** **Exp: 11/14**

Recognition Site:

5′... G T A T C C $(N)_6$ ▼ ...3′
3′... C A T A G G $(N)_5$ ▲ ...5′

Source: An *E. coli* strain that carries the cloned BciVI gene from *Bacillus circulans* (T. Le)

Supplied in: 250 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM DTT, 0.15% Triton X-100, 200 µg ml BSA and 50% glycerol.

Now Recombinant

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 C
250 mM NaCl, 10 mM Tris-HCl (pH 7.4 @ 25°C), 0.1 m M EDTA, 1 mM dithiothreitol, 0.15% Triton X-100, 200 µg/ml BSA and 50% glycerol.

Quality Control Assays

Ligation: After 10-fold overdigestion with BciVI, < 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 30 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 10 units of enzyme with 1 µg sonicated ^{3}H DNA (2 x 10^5 cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.2% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	100%
NEBuffer 2	50%
NEBuffer 3	0%
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: 65°C for 20 minutes.

Notes: BciVI produces DNA fragments that have a single-base 3′ extension which are more difficult to ligate than blunt-ended fragments.

Not sensitive to *dam*, *dcm* or mammalian CpG methylation.

Ligation was achieved using the Quick Ligation™ Kit (NEB #M2200), which contains 15% Polyethylene glycol (PEG 6000).

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

CERTIFICATE OF ANALYSIS

BciVI
NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com
R0596S 014121114111
R0596S
NEB4 37° Yes
200 units 10,000 U/ml Lot: 0141211
RECOMBINANT Store at –20°C Exp: 11/14
Recognition Site:
5′... G T A T C C (N)6 ▼ ...3′
3′... C A T A G G (N)5 ▲ ...5′
Source: An E. coli strain that carries the cloned BciVI gene from Bacillus circulans (T. Le)
Supplied in: 250 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM DTT, 0.15% Triton X-100, 200 µg ml BSA and 50% glycerol.
Now Recombinant
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 C
250 mM NaCl, 10 mM Tris-HCl (pH 7.4 @ 25°C), 0.1 m M EDTA, 1 mM dithiothreitol, 0.15% Triton X-100, 200 µg/ml BSA and 50% glycerol.
Quality Control Assays
Ligation: After 10-fold overdigestion with BciVI, < 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 30 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 10 units of enzyme with 1 µg sonicated 3H DNA (2 x 10^5 cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.2% radioactivity.
Enzyme Properties
Activity in NEBuffers:
NEBuffer 1 100%
NEBuffer 2 50%
NEBuffer 3 0%
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: 65°C for 20 minutes.
Notes: BciVI produces DNA fragments that have a single-base 3′ extension which are more difficult to ligate than blunt-ended fragments.
Not sensitive to dam, dcm or mammalian CpG methylation.
Ligation was achieved using the Quick Ligation™ Kit (NEB #M2200), which contains 15% Polyethylene glycol (PEG 6000).
= Time-Saver™ Qualified (See www.neb.com for details).
CERTIFICATE OF ANALYSIS