PshAI

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

R0593S 002121014101

R0593S

NEB4 BSA 37° Yes

1,000 units **10,000 U/ml** **Lot: 0021210**

RECOMBINANT **Store at –20°C** **Exp: 10/14**

Recognition Site:

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

Source: An *E. coli* strain that carries the cloned PshAI gene from *Plesiomonas shigelloides* (T. Shimada)

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 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 37°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 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 dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 20-fold overdigestion with PshAI, > 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 100 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction produced in one hour with one unit of enzyme.

Exonuclease Activity: Incubation of 200 units for 4 hours at 37°C in 50 µl assay buffer with 1 µg sonicated [^{3}H] DNA (10^5 cpm/µg) released < 0.1% of the radioactivity.

Endonuclease Activity: Incubation of 100 units of enzyme with 1 µg pUC 19 DNA for 4 hours at 37°C in a 50 µl reaction buffer resulted in < 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
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: A minimum of 1.0 unit is required to digest 1 µg of substrate DNA in 16 hours at 37°C.

At 25°C a minimum of 0.25 unit is required to cut 1 µg of substrate DNA in 16 hours.

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

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

For reactions longer than one hour, incubation at 25°C is recommended for best results.

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

CERTIFICATE OF ANALYSIS

PshAI

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

R0593S 002121014101

R0593S

NEB4 BSA 37° Yes

1,000 units 10,000 U/ml Lot: 0021210

RECOMBINANT Store at –20°C Exp: 10/14

Recognition Site:

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

Source: An *E. coli* strain that carries the cloned PshAI gene from *Plesiomonas shigelloides* (T. Shimada)

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 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 37°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 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 dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 20-fold overdigestion with PshAI, > 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 100 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction produced in one hour with one unit of enzyme.

Exonuclease Activity: Incubation of 200 units for 4 hours at 37°C in 50 µl assay buffer with 1 µg sonicated [^{3}H] DNA (10^5 cpm/µg) released < 0.1% of the radioactivity.

Endonuclease Activity: Incubation of 100 units of enzyme with 1 µg pUC 19 DNA for 4 hours at 37°C in a 50 µl reaction buffer resulted in < 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
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: A minimum of 1.0 unit is required to digest 1 µg of substrate DNA in 16 hours at 37°C.

At 25°C a minimum of 0.25 unit is required to cut 1 µg of substrate DNA in 16 hours.

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

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

For reactions longer than one hour, incubation at 25°C is recommended for best results.

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

CERTIFICATE OF ANALYSIS