AsiSI

NEW ENGLAND BioLabs Inc.
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R0630S 0141211113111

R0630S

NEB4 BSA 37° Yes

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

RECOMBINANT **Store at –20°C** **Exp: 11/13**

Recognition Site:

5′... GCGAT▼CGC...3′
3′... CGC▲TAGCG...5′

Source: An *E. coli* strain that carries the cloned AsiSI gene from *Arthrobacter* species (S. Degtyarev)

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

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 of XhoI digested pXba 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).

Quality Control Assays

Ligation: After 2-fold overdigestion with AsiSI, 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, > 95% can be recut.

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	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: Suitable for an extended digestion. The enzyme is active for 8 hours.

Heat Inactivation: 100 units of enzyme were inactivated by incubation at 80°C for 20 minutes.

Note: AsiSI is an isoschizomer of SgfI.

Not blocked by *dam* methylation. Cleavage of mammalian genomic DNA is blocked by CpG methylation.

Incubations longer than 8 hours are not recommended due to star activity.

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).

U.S. Patent No. 6,514,737 B1

CERTIFICATE OF ANALYSIS

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