

SexAI



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R0605S 012121014101

R0605S



200 units **4,000 U/ml** **Lot: 0121210**

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

Recognition Site:

5'...A[▼]CCWGGT...3'
3'...TGGWCC[▲]A...5'

Single Letter Code: W = A or T

Source: An *E. coli* strain that carries the cloned SexAI gene from *Streptomyces exfoliatus* (B. Frey)

Now Recombinant

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Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 8.0), 1.0 mM EDTA, 1 mM DTT, 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 DTT
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of pBC4 DNA (dcm⁻) 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)

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Quality Control Assays

Ligation: After 10-fold overdigestion with SexAI, 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 75% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 16 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 16 units of enzyme with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

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Enzyme Properties

Activity in NEBuffers:
NEBuffer 1 100%
NEBuffer 2 75%
NEBuffer 3 50%
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 0.25 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 65°C for 20 minutes.

Note: Blocked by *dcm* methylation.

Companion Products:

dam/*dcm* Competent *E. coli*
#C2925H 20 transformation reactions
#C2925I 24 transformation reactions

U.S. Patent No. 5,354,669

CERTIFICATE OF ANALYSIS

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