SgrAI

1-800-632-7799
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R0603S 005121014101

R0603S

1,000 units **10,000 U/ml** **Lot: 0051210**
RECOMBINANT **Store at –20°C** **Exp: 10/14**

Recognition Site:

5′...CR▼CCGGYG...3′
3′...GYGGCC▲RC...5′

Single Letter Code: R = A or G, Y = C or T

Source: An *E. coli* strain that carries the cloned SgrAI gene from *Streptomyces griseus* (U. Mayr)

Supplied in: 100 mM NaCl, 25 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

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 dithiothreitol
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 dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 2-fold overdigestion with SgrAI , > 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: No non-specific cleavage was observed when 3 units were incubated in a 50 µl reaction containing 1 µg of DNA for 16 hours. However star activity may be observed after incubation of greater than 4 units of this enzyme in the same reaction.

Exonuclease Activity: Incubation of 75 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.

Endonuclease Activity: Incubation of 300 units of enzyme with 1 µg ϕX174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 10% conversion to RF II.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	100%
NEBuffer 2	50%
NEBuffer 3	10%
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.

Heat Inactivation: 65°C for 20 minutes.

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

Conditions of low ionic strength, high enzyme concentration, glycerol concentration > 5% or pH > 8.0 may result in star activity.

Incubation of > 3 units SgrAI /µg of DNA substrate is not recommended.

U.S. Patent Nos. 5,134,069; 6,048,731

CERTIFICATE OF ANALYSIS

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