ScrFI

1-800-632-7799
info@neb.com
www.neb.com

R0110S 011121114111

R0110S

1,000 units **Lot: 0111211** **Exp: 11/14**
5,000 U/ml **Store at –20°C**

Recognition Site:

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

Source: An *E. coli* strain that carries the cloned ScrFI gene from *Streptococcus cremoris* F (C. Daly)

Supplied in: 250 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 0.15% Triton X-100, 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 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 2-fold overdigestion with ScrFI, < 5% 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, < 75% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 5 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 20 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 < 0.4% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	100%
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: A minimum of 0.25 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 50 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: The recognition site for ScrFI is partially modified in most *E. coli* strains. Therefore in order to digest DNA grown in them (e.g. plasmids, phages), one must use a strain deficient in cytidine methylase.

ScrFI produces DNA fragments that have a single-base 5′ extension which are more difficult to ligate than blunt-ended fragments.

Blocked by overlapping *dcm* methylation.

Cleavage of mammalian genomic DNA is blocked by overlapping CpG methylation.

Companion Products:

dam⁻/dcm⁻ Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

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

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