

StyD4I



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



R0638S

200 units 2,500 U/ml Lot: 0041208
RECOMBINANT Store at -20°C Exp: 8/14

Recognition Site:

5'...**CCNGG**...3'
3'...**GGNCC**...5'

Source: An *E. coli* strain that carries the cloned StyD4I gene from *Salmonella typhi* D4 (E.S. Anderson)

Supplied in: 300 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM dithiothreitol, 500 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:

10X NEBuffer 2.

Reaction Conditions:

1X NEBuffer 2.
Incubate at 37°C.

1X NEBuffer 2:

50 mM NaCl
10 mM Tris-HCl
10 mM MgCl₂
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 B
300 mM NaCl, 10 mM Tris-HCl, 0.1 mM EDTA,
1 mM DTT, 500 µg/ml BSA and 50% glycerol
(pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 2-fold overdigestion with StyD4I, > 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, approximately 50% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 20 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 50 units of enzyme with 1 µg sonicated [³H] DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.17% radioactivity.

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

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	10%
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 0.125 unit is required to digest 1 µg of substrate DNA in 16 hours.

Note: StyD4I is a neoschizomer of ScrFI.

Blocked by *dcm* methylation. Cleavage of mammalian genomic DNA is impaired 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).

CERTIFICATE OF ANALYSIS

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