

MboI



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



R0147S 078120714071

R0147S



500 units 5,000 U/ml Lot: 0781207

RECOMBINANT Store at -20°C Exp: 7/14

Recognition Site:

5'...GATC...3'
3'...CTAG...5'

Source: An *E. coli* strain that carries the cloned MboI gene from *Moraxella bovis* (ATCC 10900)

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

New Reaction Buffer

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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 (*dam*⁻) 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 10-fold overdigestion with MboI, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5' termini concentration

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of 1–2 µM) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 100 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 100 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.

Endonuclease Activity: Incubation of 100 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	75%
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.

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NEBuffer 1	75%
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Survival in a Reaction: A minimum of 0.50 unit is required to digest 1 µg of substrate DNA in 16 hours.

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

Notes: BfuCI, DpnII and Sau3AI are isoschizomers of MboI. MboI cleaves to leave a 5' GATC extension which can be efficiently ligated into BamHI, BclII, BglII, BstYI, DpnII or Sau3AI cleaved fragments.

Blocked by *dam* methylation. It's isoschizomer Sau3AI is not.

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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