



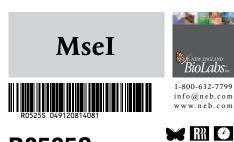
500 units 10,000 U/ml Lot: 0491208 RECOMBINANT Store at -20°C Exp: 8/14

Recognition Site:

5′... T^VT A A ... 3′ 3′... A A T_AT ... 5′

Source: An *E. coli* strain that carries the cloned Msel gene from *Micrococcus species* (R. Morgan)

New Reaction Buffer





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NEB 4 BSA 37° Yasa

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5′... T^VT A A ... 3′ 3′... A A T_AT ... 5′

Source: An *E. coli* strain that carries the cloned Msel gene from *Micrococcus species* (R. Morgan)

Supplied in: 50 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 200 μ g/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:

25°C)

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 μ g of λ DNA in 1 hour at 37°C in a total reaction volume of 50 μ l.

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 @

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Quality Control Assays

Ligation: After 10-fold overdigestion with Msel, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5´ termini concentration of 1–2 μ M) at 16°C. Of these ligated fragments, > 95% can be recut.

16-Hour Incubation: A 50 μ I reaction containing 1 μ g 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 μ g sonicated ³H DNA (10⁵ cpm/ μ g) for 4 hours at 37°C in 50 μ l reaction buffer released < 0.4% radioactivity.

<u>Enzyme Properties</u>

Activity in NEBuffers:

 NEBuffer 1
 75%

 NEBuffer 2
 100%

 NEBuffer 3
 75%

 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 0.13 unit is required of digest 1 μ g of substrate DNA in 16 hours.

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

Note: Not sensitive to *dam, dcm* or mammalian CpG methylation.

Image: Saver[™] Qualified (See www.neb.com for details).



CERTIFICATE OF ANALYSIS

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Enzyme Properties

Activity in NEBuffers:

NEBuffer 175%NEBuffer 2100%NEBuffer 375%

NEBuffer 4 **100**%

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