



R0538S

1,000 units	10,000 U/ml	Lot: 0021211
RECOMBINANT	Store at -20°C	Exp: 11/14

100

BioLabs

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NEB 4 37° 😪

Recognition Site:

5′...3′ 3′... T T A A ...5′

Source: An *E. coli* strain that carries the cloned MluCl gene from *Micrococcus luteus*

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.



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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.

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 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 KCI, 10 mM Tris-HCI, 0.1 mM EDTA, 1 mM DTT, 200 μg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Heat Inactivation: 80°C for 20 minutes.

Quality Control Assays

Ligation: After 20-fold overdigestion with MluCI, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at 5^{\prime} termini concentration 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 30 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 μ g sonicated [³H] DNA (10⁵ cpm/ μ g) for 4 hours at 37°C in 50 μ l reaction buffer released < 0.5% radioactivity.

Enzyme Properties

Activity in NEBuffers:

 NEBuffer 1
 100%

 NEBuffer 2
 10%

 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.

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

Notes: MluCl is an isochizomer of Tsp5091.

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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