



R0197S

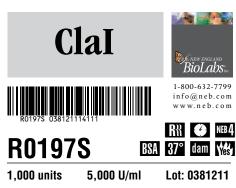
1,000 units 5,000 U/ml Lot: 0381211 RECOMBINANT Store at -20°C Exp: 11/14

Recognition Site:

5′... A T^VC G A T ... 3′ 3′... T A G C T A ... 5′

Source: An *E. coli* strain that carries the cloned Clal gene from *Caryophanon latum* L (ATCC 49862)

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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Reagents Supplied with Enzyme: 10X NEBuffer 4, 100X BSA.

Reactions Conditions: 1X NEBuffer 4, supplemented with 100 µg/ml BSA. Incubate at 37°C.

1X NEBuffer 4:

BioLabs

1-800-632-7799

info@neb.com

www.neb.com

R 🕻 🕐 NEB 4

BSA 37° dam 🐝

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 (*dam*⁻) 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 dithiothreitol, 200 μg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

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Quality Control Assays

Ligation: After 10-fold overdigestion with Clal, > 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 200 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 250 units of Clal with 1 μ g sonicated ³H DNA (10⁵ cpm/ μ g) for 4 hours at 37°C in 50 μ l reaction buffer released < 0.1% radioactivity.

Endonuclease Activity: Incubation of 30 units of Clal with 1 μ g ϕ X174 RF I DNA for 4 hours at 37°C in 50 μ l reaction buffer resulted in < 10% conversion to RF II.

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

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Enzyme Properties

Activity in NEBuffers:

 NEBuffer 1
 10%

 NEBuffer 2
 50%

 NEBuffer 3
 50%

 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.

Heat Inactivation: 65°C for 20 minutes.

Plasmid Cleavage: Number of units required to cleave 1 μ g supercoiled plasmid DNA in one hour: pBR322 = 5 units.

Notes: Clal is an isoschizomer of BspDI.

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

Companion Products:

dam-/dcm- Competent E. coli#C2925H20 transformation reactions#C2925I24 transformation reactions

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 CERTIFICATE OF ANALYSIS

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