AlwI













500 units 5.000 U/ml RECOMBINANT Store at -20°C Exp: 7/14

Lot: 0181207

Recognition Site:

5′... G G A T C (N)₄ ▼... 3′ 3′... C C T A G (N)₅,...5′

Source: An E. coli strain that carries the cloned Alwl gene from *Acinetobacter Iwoffii* (R. Morgan)

More Units



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 µg of λ DNA (dam -) in 1 hour at 37°C in total reaction volume of 50 ul.

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 @ 25°C)

Quality Control Assays

Ligation: After 2-fold overdigestion with Alwl, approximately 50% 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, > 95% can be recut.

16-Hour Incubation: A 50 ul reaction containing 1 µg of DNA and 5 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 25 units of enzyme with 1 μg sonicated [3H] DNA (105 cpm/μg) for 4 hours at 37°C in 50 ul reaction buffer released < 0.1% radioactivity.

Enzyme Properties Activity in NEBuffers

NEBuffer 1 50% NEBuffer 2 100% 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.

Survival in a Reaction: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 25 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: AlwI produces DNA fragments that have a single-base 5' extension which are more difficult to ligate than blunt-ended fragments.

Blocked by dam methylation.

= Time-Saver™ Qualified (See www.neb.com for details).

CERTIFICATE OF ANALYSIS

AlwI



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

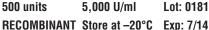












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