

# AseI



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



R0526S 012120614061

## R0526S



**2,000 units**    **10,000 U/ml**    **Lot: 0121206**  
**RECOMBINANT**    **Store at -20°C**    **Exp: 6/14**

### Recognition Site:

5'...ATTAAAT...3'  
3'...TAAATTA...5'

**Source:** An *E. coli* strain that carries the cloned AseI gene from *Aquaspirillum serpens* (ATCC 12638)

Supplied in: 500 mM KCl, 10 mM Tris-HCl (pH 7.5), 1 mM DTT, 0.1 mM EDTA, 200 µg/ml BSA and 50% glycerol.

**Reagents Supplied with Enzyme:**  
10X NEBuffer 3.

**Reaction Conditions:** 1X NEBuffer 3.  
Incubate at 37°C.

**1X NEBuffer 3:**  
100 mM NaCl  
50 mM Tris-HCl  
10 mM MgCl<sub>2</sub>  
1 mM DTT  
pH 7.9 @ 25°C

**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1 µg of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

**Diluent Compatibility:** Diluent Buffer B  
300 mM NaCl, 10 mM Tris-HCl, 0.1 mM EDTA,  
1 mM DTT, 500 µg/ml BSA and 50% glycerol  
(pH 7.4 @ 25°C).

### Quality Control Assays

**Ligation:** After 20-fold overdigestion with AseI, > 95% 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 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 300 units of enzyme with 1 µg sonicated <sup>3</sup>H DNA (10<sup>5</sup> cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

### Enzyme Properties

#### Activity in NEBuffers:

NEBuffer 1    NR  
NEBuffer 2    75%  
NEBuffer 3    **100%**  
NEBuffer 4    NR

NEBuffer 2 may be used for incubations less than 1 hour. NEBuffers 1 and 4 are **not** recommended (NR) due to star activity.

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 to digest 1 µg of substrate DNA in 16 hours.

**Heat Inactivation:** 12 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

**Plasmid Cleavage:** Number of units required to cleave 1 µg of supercoiled plasmid DNA in one hour: pBR322 = 0.3 unit.

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

AseI cleaves pBR322 and adenovirus-2 DNA at a rate 3 times that of λ DNA.

Conditions of low ionic strength, high enzyme concentration, glycerol concentration > 5% or pH > 8.0 may result in star activity.

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

CERTIFICATE OF ANALYSIS

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