





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

R0108S



3.000 units RECOMBINANT Store at -20°C Exp: 11/14

10.000 U/ml Lot: 0321211

Recognition Site:

5′... G G C C ... 3′ 3′... C C₄G G ... 5′

Source: An *E. coli* strain that carries the cloned HaellI gene from *Haemophilus aegypticus* (ATCC 11116)

New Reaction Buffer

Supplied in: 100 mM KCI, 10 mM Tris-HCI (pH 7.4), 0.1 mM EDTA, 10 mM 2-mercaptoethanol, 500 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 μa of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A 50 mM KCI, 10 mM Tris-HCI, 0.1 mM EDTA, 1 mM DTT. 200 ug/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 20-fold overdigestion with HaellI. > 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 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 100 units of enzyme with 1 ug sonicated 3H DNA (105 cpm/ μg) for 4 hours at 37°C in 50 μl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1 50% NFBuffer 2 100% NEBuffer 3 25% 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 0.25 unit is required to digest 1 ug of substrate DNA in 16 hours.

Heat Inactivation: 80°C for 20 minutes

Notes: GGCm5C is cleaved, but GGm5CC is resistant to cleavage.

Not sensitive to dam, dcm or mammalian CpG methylation.

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

CERTIFICATE OF ANALYSIS

HaeIII



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1X NEBuffer 4:

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Quality Control Assays

Ligation: After 20-fold overdigestion with Haelll, > 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 ul reaction containing 1 µg 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.

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