

NheI



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



R0131S 027121014101

R0131S



1,000 units 10,000 U/ml Lot: 0271210
RECOMBINANT Store at -20°C Exp: 10/14

Recognition Site:

5'... GCTAGC... 3'
3'... CGATCG... 5'

Source: An *E. coli* strain that carries the cloned *NheI* gene from *Neisseria mucosa heidelbergensis* (ATCC 25999)

Also Available In
High-Fidelity (HF™) Format

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Supplied in: 250 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 0.15% Triton X-100, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 2, 100X BSA

Reaction Conditions: 1X NEBuffer 2, supplemented with 100 µg/ml BSA. Incubate at 37°C.

1X NEBuffer 2:
50 mM NaCl
10 mM Tris-HCl
10 mM MgCl₂
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 (HindIII digest) in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer C
250 mM NaCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 0.15% Triton X-100, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C).

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Quality Control Assays

Ligation: After 100-fold overdigestion with *NheI*, > 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 60 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 700 units of enzyme with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Endonuclease Activity: Incubation of 30 units of enzyme with 1 µg φX174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 10% conversion to RF II.

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Enzyme Properties

Activity in NEBuffers:
NEBuffer 1 100%
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: A minimum of 0.25 unit is required to digest 1 µg of substrate DNA in 16 hours.

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

Notes: Cleaves to leave a 5' CTAG extension which can be efficiently ligated to DNA fragments generated by *AvrII*, *SpeI* or *XbaI*.

Cleavage of mammalian genomic DNA is blocked by some combinations of overlapping CpG methylation.

Activity inhibited by salt concentrations > 100 mM.

(See other side)

CERTIFICATE OF ANALYSIS

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Companion Products Sold Separately:

Nhel-HF™
#R3131S 1,000 units
#R3131L 5,000 units
#R3131M 5,000 units

Nhel-HF™ RE-Mix™
#R5131S 50 reactions

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

U.S. Patent No. 6,387,681

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#R3131L 5,000 units
#R3131M 5,000 units

Nhel-HF™ RE-Mix™
#R5131S 50 reactions

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U.S. Patent No. 6,387,681