



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

R3131S

NEB 4

BSA

37°

Yes


1,000 units 20,000 U/ml Lot: 0021210
RECOMBINANT Store at -20°C Exp: 10/14


Recognition Site:

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

Note: NheI-HF has the same specificity as NheI (NEB #R0131), but it has been engineered for reduced star activity.

Source: An *E. coli* strain that carries the cloned and modified (E77A) NheI gene from *Neisseria mucosa heidelbergensis* (ATCC 25999)



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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 4, 100X BSA

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

1X NEBuffer 4:
50 mM potassium acetate
20 mM Tris-acetate
10 mM magnesium acetate
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 (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
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Quality Controls

Ligation: After 100-fold overdigestion with NheI-HF, > 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 200 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 100 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 10%
NEBuffer 3 0%
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 0.13 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 25 units of enzyme were inactivated by incubation at 80°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.

(See other side)

CERTIFICATE OF ANALYSIS

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(See other side)

CERTIFICATE OF ANALYSIS

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




Activity inhibited by salt concentrations > 100 mM.

Companion Products Sold Separately:

NheI	
#R0131S	1,000 units
#R0131L	5,000 units
#R0131M	5,000 units

NheI-HF™ RE-Mix™	
#R5131S	50 reactions

New icons (see www.neb.com for details)

-  = Time-Saver™ Qualified
-  = indicates that the enzyme has been engineered
-  = indicates that the enzyme has reduced star activity

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

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




Activity inhibited by salt concentrations > 100 mM.

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#R0131L	5,000 units
#R0131M	5,000 units

NheI-HF™ RE-Mix™	
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