

DpnII



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



R0543S 010121114111

R0543S



1,000 units 10,000 U/ml Lot: 0101211
RECOMBINANT Store at -20°C Exp: 11/14

Recognition Site:

5'...▼GATC...3'
3'...CTAG▲...5'

Source: An *E. coli* strain that carries the cloned DpnII gene from *Diplococcus pneumoniae* G41 (S. Lacks)

New Storage Conditions

Supplied in: 300 mM NaCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 500 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C).

Reagents Supplied with Enzyme:
10X NEBuffer DpnII.

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

1X NEBuffer DpnII:
100 mM NaCl
50 mM Bis Tris-HCl
10 mM MgCl₂
1 mM dithiothreitol
pH 6.0 @ 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 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 dithiothreitol, 500 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 50-fold overdigestion with DpnII, > 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 100 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.

Enzyme Properties

Activity in NEBuffers:

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

NEBuffers 1, 2 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: 50 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

(see other side)

CERTIFICATE OF ANALYSIS

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CERTIFICATE OF ANALYSIS

Notes: DpnII and Sau3AI are isoschizomers of MboI.

Cleaves to leave a 5' GATC extension which can be efficiently ligated to DNA fragments generated by BamHI, BclI, BglII, MboI, Sau3AI and BstYI.

Blocked by *dam* methylation.

DpnII exhibits star activity when incubated in a buffer with pH > 6.5.


DpnII is not recommended for use in any buffer except its own unique buffer because of the resultant lower activity and potential star activity.

Companion Products:

dam⁻/dcm⁻ Competent *E. coli*

#C2925H 20 transformation reactions

#C2925I 24 transformation reactions

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



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