

Bpu10I



1-800-632-7799
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R0649S 004120914091

R0649S

200 units **5,000 U/ml** **Lot: 0041209**

RECOMBINANT **Store at -20°C** **Exp: 9/14**

Recognition Site:

5'... CCTNAGC...3'
3'... GGANTCG...5'

Source: Two *E. coli* strains that carry the cloned subunits of Bpu10I from *Bacillus pumilus*10 (S.K. Degtyarev)

Supplied in: 300 mM NaCl, 10 mM Tris-HCl (pH 7.4 @ 25°C), 0.1 mM EDTA, 1 mM DTT, 500 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 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 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 7.5 @ 25°C).

Quality Control Assays

Ligation : After 10-fold overdigestion with Bpu10I, > 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, approximately 50% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of λ DNA and 5 units of enzyme incubated for 16 hours at 37°C resulted in a DNA pattern free of detectable nuclease degradation as determined by gel electrophoresis.

Exonuclease Activity: Incubation of a 50 µl reaction containing 25 units of Bpu10I with 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA (200,000 cpm/µg) for 4 hours at 37°C released < 0.1% of the total radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	10%
NEBuffer 2	25%
NEBuffer 3	100%
NEBuffer 4	25%

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: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 80°C for 20 minutes.

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

Not sensitive to *dam*, *dcm*, or CpG methylation.

CERTIFICATE OF ANALYSIS

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