BaeI

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

R0613S 008121013101

R0613S

250 units **Lot: 0081210** **Exp: 10/13**
5,000 U/ml **Store at –20°C**

Recognition Site:

5′..▼10(N) A C (N)4 G T A Y C (N)12▼.3′
3′..▲15(N) T G (N)4 C A T R G (N)7▲..5′

Single Letter Code: R = A or G, Y = C or T

Source: *Bacillus sphaericus* (H. Kong)

New Reaction Buffer

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 4, 100X BSA, (32 mM) SAM.

Reaction Conditions: 1X NEBuffer 4, supplemented with 100 µg/ml BSA, (20 µM) SAM. Incubate at 25°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 µg of λ DNA in 1 hour at 25°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol (pH 7.9 @ 25°C)

Quality Control Assays

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 8 units of enzyme incubated for 16 hours resulted in no degradation of the DNA bands due to nonspecific nucleases. However, fragments produced by noncanonical cleavage due to star activity may be observed with 16 units of enzyme in similar conditions.

Exonuclease Activity: Incubation of 8 units of enzyme with 1 µg sonicated ^{3}H DNA (10^{5} cpm/µg) for 4 hours at 25°C in 50 µl reaction buffer released < 0.01% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
NEBuffer 2	**100%**
NEBuffer 3	50%
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 0.5 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 65°C for 20 minutes.

Notes: BaeI cleaves DNA substrates twice to excise its recognition site generating a 28 base-pair fragment with 5 base 3′ overhangs. In one sequence context, BaeI was found to cut at its defined cleavage site as well as one base pair further from its recognition site.

Requires S-adenosylmethionine for optimal activity (supplied with enzyme).

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

Incubation at 37°C results in 20% activity.

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

CERTIFICATE OF ANALYSIS

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