Taq$^{\alpha}$I

NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com

R0149S 0541211114111

R0149S

RR NEB4 BSA 65° Yes dam

4,000 units **20,000 U/ml** **Lot: 0541211**
RECOMBINANT **Store at –20°C** **Exp: 11/14**

Recognition Site:

5′. . . T▼CGA . . . 3′
3′. . . AGC▲T . . . 5′

Source: An *E. coli* strain that carries a Taq$^{\alpha}$I overproducing plasmid (F. Barany using an NEB clone). Taq$^{\alpha}$I has a two amino acid replacement at its amino terminus.

New Reaction Buffer

Supplied in: 300 mM KCl, 10 mM Tris-HCl (pH 7.5), 1 mM EDTA, 1 mM DTT, 500 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 65°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 65°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 7.4 @ 25°C).

Quality Control Assays

Ligation: After 50-fold overdigestion with Taq$^{\alpha}$I, > 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 fragment, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 300 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 500 units of enzyme with 1 µg sonicated ^{3}H DNA (10^{5} cpm/µg) for 4 hours at 65°C in 50 µl reaction buffer released < 0.12% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
NEBuffer 2	75%
NEBuffer 3	100%
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 0.50 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 100 units of enzyme were inactivated by incubation at 80°C for 20 minutes.

Notes: Taq$^{\alpha}$I is modified as described above.

Blocked by overlapping *dam* methylation.

Incubation without BSA results in 50% activity. Incubation at 37°C results in 10% activity. Cleaves single-stranded DNA slowly.

Companion Products:
dam^{-}/dcm^{-} Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

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

CERTIFICATE OF ANALYSIS

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