



NEB 4 37°

in fo@neb.com www.neb.com

# R0710S

250 units 5,000 U/ml Lot: 0011208 RECOMBINANT Store at -20°C Exp: 8/14

#### **Recognition Site:**

5´... R G<sup>V</sup>C Y ... 3´ 3´... Y C<sub>A</sub>G R ... 5´

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

**Description:** CviKI-1 is a restriction endonuclease with 4 expected recognition sites as well as up to eleven relaxed non-cognate sites (star sites).



Supplied in: 50 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

**Reagents Supplied with Enzyme:** 10X NEBuffer 4.

**Reaction Conditions:** 1X NEBuffer 4. 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  $\mu$ g of pBR322 DNA in 1 hour at 37°C in a total reaction volume of 50  $\mu$ l.

### Diluent Compatibility: Diluent Buffer A

50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200  $\mu g/ml$  BSA and 50% glycerol (pH 7.4 @ 25°C)

# **Quality Control Assays**

**Ligation:** After 10-fold overdigestion with CviKI-1, > 95% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5' termini concentration of  $1-2 \ \mu$ M) at 16°C. Of these ligated fragments, > 95% can be recut.

**16-Hour Incubation:** A 50  $\mu$ I reaction containing 1  $\mu$ g of DNA and 10 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.

# Enzyme Properties

Activity in NEBuffers NEBuffer 1 10% NEBuffer 2 50% NEBuffer 3 25% 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 1 unit is required to digest 1  $\mu g$  of substrate DNA in 16 hours.

Heat Inactivation: 80°C for 20 minutes.

**Notes:** CviKI-1 overnight digestion or addition of 20% DMSO greatly enhances the star activity. DNA can be digested to small oligos under "star" conditions.

■ Time-Saver<sup>™</sup> Qualified (See www.neb.com for details).

CERTIFICATE OF ANALYSIS



**Recognition Site:** 

5<sup>'</sup>... R G<sup>V</sup>C Y ... 3<sup>'</sup> 3<sup>'</sup>... Y C<sub>A</sub>G R ... 5<sup>'</sup> Single Letter Code: R = A or G, Y = C or T

**Description:** CviKI-1is a restriction endonuclease with 4 expected recognition sites as well as up to eleven relaxed non-cognate sites (star sites).

**Source:** An *E. coli* strain carrying the cloned and modified CviKI-1 restriction gene derived from CA-1A, a *Chlorella* virus.

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