## EciI





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

## **R0590S**



50 units 2 000 H/ml Lot: 0181211 Exp: 11/13

2,000 U/ml Store at -20°C

### **Recognition Site:**

5'...GGCGGA(N)<sub>11</sub>...3' 3'...CCGCCT(N)<sub>9</sub>...5'

**Source:** An *E. coli* strain that carries the cloned Ecil gene from *Escherichia coli* T-8 (R. Croft)

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

### **Now Recombinant**

### 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 dithiothreitol pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1  $\mu g$  of  $\lambda$  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 2-fold overdigestion with Ecil, approximately 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.

**Exonuclease Activity:** Incubation of 2 units of enzyme with 1  $\mu$ g sonicated <sup>3</sup>H DNA (10<sup>5</sup> cpm/ $\mu$ g) for 4 hours at 37°C in 50  $\mu$ l reaction buffer released < 0.1% radioactivity.

### **Enzyme Properties**

### Activity in NEBuffers:

NEBuffer 1 100% NEBuffer 2 50% 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 1.0 unit is required to digest 1  $\mu g$  of substrate DNA in 16 hours.

**Heat Inactivation:** 65°C for 20 minutes.

**Notes:** Activity in NEBuffer 2 can be increased to 100% with the addition of 100 µg/ml BSA.

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

Incubations longer than 4 hours are not recommended.

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

CERTIFICATE OF ANALYSIS

## **EciI**



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

# \_\_\_\_



# **R0590S**



50 units Lot: 0181211 Exp: 11/13 2.000 U/ml Store at -20°C

### **Recognition Site:**

 $5' \dots G G G G A (N)_{11}^{\Psi} \dots 3'$  $3' \dots G G G G T (N)_{9_{\underline{A}}} \dots 5'$ 

**Source:** An *E. coli* strain that carries the cloned Ecil gene from *Escherichia coli* T-8 (R. Croft)

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

**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 dithiothreitol pH 7.9 @ 25°C

**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1  $\mu$ g of  $\lambda$  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 2-fold overdigestion with Ecil, approximately 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.

**Exonuclease Activity:** Incubation of 2 units of enzyme with 1  $\mu$ g sonicated <sup>3</sup>H DNA (10<sup>5</sup> cpm/ $\mu$ g) for 4 hours at 37°C in 50  $\mu$ l reaction buffer released < 0.1% radioactivity.

#### **Enzyme Properties**

### Activity in NEBuffers:

NEBuffer 1 100% NEBuffer 2 50% 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 1.0 unit is required to digest 1  $\mu g$  of substrate DNA in 16 hours.

Heat Inactivation: 65°C for 20 minutes.

**Notes:** Activity in NEBuffer 2 can be increased to 100% with the addition of 100  $\mu$ g/ml BSA.

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

Incubations longer than 4 hours are not recommended.

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

**Now Recombinant**