

Eco53kl



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



R0116S 004120714071

R0116S



1,000 units **10,000 U/ml** **Lot: 0041207**

RECOMBINANT **Store at -20°C** **Exp: 7/14**

Recognition Site:

5'...GAG ∇ CTC...3'
3'...CTC \blacktriangle GAG...5'

Source: An *E. coli* strain that carries the cloned Eco53kl gene from *Escherichia coli* 53k (A.S.Solonin)

Supplied in: 50 mM KCl, 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 µg of pXba DNA in 1 hour at 37°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 7.4 @ 25°C).

Quality Control Assays

Ligation: After 10-fold overdigestion with Eco53kl, approximately 75% 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, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg 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.

Exonuclease Activity: Incubation of 100 units of enzyme with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	100%
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: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 65°C for 20 minutes.

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

CERTIFICATE OF ANALYSIS

Eco53kl



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



R0116S 004120714071

R0116S



1,000 units **10,000 U/ml** **Lot: 0041207**

RECOMBINANT **Store at -20°C** **Exp: 7/14**

Recognition Site:

5'...GAG ∇ CTC...3'
3'...CTC \blacktriangle GAG...5'

Source: An *E. coli* strain that carries the cloned Eco53kl gene from *Escherichia coli* 53k (A.S.Solonin)

Supplied in: 50 mM KCl, 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 µg of pXba DNA in 1 hour at 37°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 7.4 @ 25°C).

Quality Control Assays

Ligation: After 10-fold overdigestion with Eco53kl, approximately 75% 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, > 95% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg 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.

Exonuclease Activity: Incubation of 100 units of enzyme with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	100%
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: Intermediate activity. Suitable for extended digestion, but < 8 hours.

Heat Inactivation: 65°C for 20 minutes.

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

CERTIFICATE OF ANALYSIS