

Product Information

RESTRICTION ENDONUCLEASE EcoR I

Product No. **R 4640**

Store at 0 to -20 °C

Product Summary

Recognition Sequence: 5' G/AATTC'3

Activity: 40,000 units/ml

Cutting: 100%

Ligation: >95%

Recutting: >95%

No degradation detected with >100 units for 16 hrs.

Fold over digestion: 1,600 (100 units x 16 hrs.)

Package Size: 1,000 units and 5,000 units

Unit Definition

One unit is the enzyme activity that completely cleaves 1 µg λDNA in 1 hr. at 37 °C in a total volume of 50 µl of Buffer SH for restriction enzymes. 1 µg pBR322 DNA is digested completely by 2 units of Eco RI.

Specificity

Eco RI recognizes the sequence G/AATTC and generates fragments with 5'-cohesive termini.¹ Eco RI is an isoschizomer to Rsa I. Eco RI is inhibited by the presence of N⁶-methyladenine at either or both A residues in the sequence G^mA^mATTC.

Comments

Digestion Buffer SH is supplied as a 10x concentrate.

0.5-100 units of Eco RI is not heat inactivated after incubation at 65 °C for 15 min.

Eco RI Storage and Dilution Buffer Composition

10 mM Tris-HCl

200 mM NaCl

1.0 mM EDTA

0.5 mM dithioerythritol

0.2% Triton[®] X-100

50% (v/v) glycerol

pH 7.0

1x Digestion Buffer SH (B 3657) Composition for

Eco RI: 100% Digestion at 37 °C.

50mM Tris-HCl

100 mM NaCl

10 mM MgCl₂

1 mM dithioerythritol

pH 7.5

Quality Control Testing

Absence of unspecific endonuclease activities:

1 µg λDNA is incubated for 16 hrs. in 50 µl buffer SH with >100 units of Eco RI.

Ligation and Recutting Assay

Eco RI fragments obtained by complete digestion of 1 µg λDNA are adjusted to pH 7.5 at 20 °C. The Eco RI fragments are then ligated with 0.1 unit T4-DNA ligase at pH 7.5 at 4 °C. A 10 µl reaction mixture, incubated for 16 hrs. at 4 °C, contained: 0.1 units T4-DNA ligase, 66 mM Tris-HCl, 5 mM MgCl₂, 1 mM dithioerythritol, and 1 mM ATP.

The degree of ligation and subsequent recutting with Eco RI to yield the typical pattern of λxEcoRI fragments is determined.

References

1. Hedgpeth, J., et al., Proc. Natl. Acad. Sci USA, **69**, 3448 (1972).

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.