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Certificate of Analysis

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EcoR V

Catalog No: 2160

Lot No: 2307033

Package Size: See Product Label

Concentration: 10 units/ μ l

Source: *Escherichia coli* J62plg74

Storage Conditions: Store at -20°C

Recognition/Cut Site

5'-GAT↓ATC-3'

3'-CTA↑TAG-5'

Unit Definition

One unit is the amount of enzyme required to completely digest 1 μ g of lambda DNA in 1 hour at 37°C.

Heat Inactivation

10 minutes at 80°C

Reaction Temperature

37°C

Reaction Conditions

50 mM Tris-HCl (pH 7.5 at 37°C)

100 mM NaCl

10 mM MgCl₂

1 mM dithiothreitol (DTT)

100 μ g/ml bovine serum albumin*

*supplied separately

Storage Buffer

10 mM Tris-HCl (pH 7.4)

50 mM KCl

0.1 mM EDTA

1 mM dithiothreitol (DTT)

0.1% Triton X-100™

200 μ g/ml bovine serum albumin

50% (v/v) glycerol

Assay Conditions

50 mM Tris-HCl (pH 7.5 at 37°C)

10 mM MgCl₂

100 mM NaCl

1 mM dithiothreitol (DTT)

0.025% Triton X-100™

100 μ g/ml bovine serum albumin

1 μ g Lambda DNA

Incubation time is 1 hour at 37°C

Reaction volume of 50 μ l

Quality Control

Non-specific Endonuclease: 20, 35, and 50 units of EcoR V were incubated with Lambda DNA for 5 hours at 37°C and compared to a standard 4 unit, 1 hour assay. A second assay was performed under the same conditions using pBR322 as the DNA.

3'-Exonuclease: 5, 10, 20, and 30 units of EcoR V were incubated with [³H] labeled 3'-ends of Lambda/Taq I DNA fragments for 1 hour at 37°C.

5'-Exonuclease/5'-Phosphatase: 5, 10, 20, and 30 units of EcoR V were incubated with [γ - ³²P] labeled 5'-ends of Lambda/Hae III DNA fragments for 1 hour at 37°C.

Nicking: 20, 35, and 50 units of EcoR V were incubated with Φ X-174 DNA for 5 hours at 37°C.

Ligation/Recut: 4 units of EcoR V per μ g of Lambda DNA were incubated for 1 hour at 37°C. Resulting fragments were ligated with T4 DNA Ligase and recut with EcoR V.