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ProductInformation

RESTRICTION ENDONUCLEASE Hpa II

Product No. **R 0629**

Store at 0 °C to –20 °C

Product Summary
Recognition Sequence: 5'C/CGG'3
Activity: 10,000 units/ml
Cutting: 100%
Ligation: >95%
Recutting: >95%
No degradation detected with >40 units for 16 hrs.
Fold over digestion: 640 (40 units x 16 hrs.)
Package Size: 500 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 25 µl of Buffer SL for restriction endonucleases.

Specificity
Hpa II recognizes the sequence C/CGG and generates fragments with 5'-cohesive termini.[1] Hpa II is sensitive to methylation at the internal cytosine. Hpa II does not cleave ^{m}CCGG and C^{m}CGG .

Comments
Digestion Buffer SL is supplied as a 10x concentrate. Information for heat inactivation Hpa II is not available.

Hpa II Storage and Dilution Buffer Composition
20 mM Tris-HCl
50 mM KCl
0.5 mM EDTA
5 mM 2-mercaptoethanol
50% (v/v) glycerol
pH 7.5

1x Digestion Buffer SL (B 3782) Composition for Hpa II: 100% Digestion at 37 °C.
10 mM Tris-HCl
10 mM $MgCl_2$
1 mM dithioerythritol (DTE)
pH 7.5

Quality Control Testing
Absence of unspecific endonuclease activities:
1 µg λDNA is incubated for 16 hrs. in 50 µl buffer SL with excess of Hpa II.

Ligation and Recutting Assay
Hpa II fragments, obtained by complete digestion of 1 µg λ DNA, are adjusted to pH 7.5 at 20 °C. The Hpa II fragments are then ligated with 0.5 unit T4-DNA ligase at pH 7.5 at 20 °C. A 10 µl reaction mixture, incubated for 16 hrs. at 20 °C, contained 0.5 unit T4-DNA ligase, 66 mM Tris-HCl, 5 mM $MgCl_2$, 1mM dithiothreitol and 1 mM ATP.

The degree of ligation and subsequent recutting with Hpa II to yield the typical pattern of λ x Hpa II fragments is determined.

References
1. Garfin, D., et al., Biochem. Biophys. Res. Commun., **59**, 108 (1974).

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