EcoR V

GAT^ATC AccuCut^{IM} Restriction Endonuclease

• Cat. No. E-1631 2000 Units E-1632 10000 Units

Lot No.: 02I131491H8A3

Supplied with Enzyme

10X AccuCut™Blue Buffer : 1 mL 100 mM pH 8.5 Tris-HCI 100 mM MaCl₂ 1 M NaCl 10 mM DTT 1X Dilution Buffer : 1 mL 10 mM pH 7.6 Tris-HCI 50 mM KCI $0.1 \, \text{mM}$ **FDTA** 1 mM DTT $200 \mu \text{ g/mL}$ Acetylated BSA 50% Glycerol

- Store at -20 C.
- Unit definition : One unit of restriction endonuclease activity is defined as the amount of enzyme required to completely digest 1µg of substrate DNA in a total reaction volume of 50 µL in one hour using the AccuCut™ buffer provided. Incubations are performed in 1.5 mL tubes at the appropriate incubation temperature as indicated in the Product Profile.

• Isoschizomer : Ceg | ,Eco32 |.

· Neoschizomer: Unfound

- Reactivity on methylated substrate DNA: Blocked by G m6ATATC, GAT m6ATC. Not blocked by GATAT m6C, GATAT m6C.
- Ref) 1.Nwosu, V.U., Connolly, B.A., Halford, S.E., Garnett, J., (1988) Nucleic Acids Res., vol. 16, pp. 3705-3720. 2.Schildkraut, I., Banner, C.D.B., Rhodes, C.S., Parekh, S., (1984) Gene, vol. 27, pp. 327-329. 3.Vipond, I.B., Moon, B.J., Halford, S.E., (1994) Biochem. Soc. Trans., vol. 22, pp. 301S.

· Source : Escherichia coli.

• Concentration : 15 Units/µL

Reaction Condition

- 10X AccuCut™ Blue Buffer

Incubate at 37℃.

Storage Buffer

20 mM pH 7.5, Tris-HCl 50 mM KCl 1 mM EDTA

10 mM 2-mercaptoethanol

50% Glycerol

• Heat inactivation : No.

Quality Control

• Overdigestion Assay :

No nonspecific activity was detected after incubation of 1 μg of λ DNA with 50 units of *EcoR* V for 15 hours.

* Conditions of low ionic strength, high enzyme concentration, glycerol concentration >5%, or pH >8.0 may result in star activity.

· Nuclease Contamination Assay :

No altered pattern was detected after incubation of 1 µg of substrate DNA with *Eco*R V in 50 µL reaction volume with the supplied AccuCut™ buffer overnight.

. Ligation and Recutting Assay:

This assay is used to test for exonuclease activity that would degrade the termini of restriction fragments, resulting

in inhibition of ligation and of subsequent digestion of ligated fragments. After 40-fold overdigestion with *EcoR V*, 95% of the DNA fragments can be ligated and recut with *EcoR V*.