

Restriction Enzyme EcoR V



Cat # FG-EcoRV

Size 4.000 units

Conc. 20 units/ul

Store at -20°C

Supplied with: 10X FastGene® Buffer III (FG-REB3)

10X FastGene® FastCut Buffer (FG-REBHF)

6X DNA Loading Buffer

Sterile water

Recognition site

For Research Use Only. Not for use in diagnostic procedures.

ISO9001

Dilution buffer

FastGene® Diluent A

Heat Inactivation

EcoR V can be inactivated at 80°C for 20 min.

Methylation sensitivity

dam methylation: Not sensitive dcm methylation: Not sensitive

CpG methylation: Conditionally sensitive

Prolonged incubation

A minimum amount of enzyme required to digest 1 µg substrate DNA for 16 hr; 0.5 U.

Relative activity in FastGene® Buffers

FastGene® Buffer I: FastGene® Buffer II: 100% FastGene® Buffer III: 100% FastGene® Buffer IV: 50% FastGene® FastCut Buffer: 100%

Note

No further activity is noted after 4 hr incubation at 37°C and thus overnight incubation is not effective. Cleavage of mammalian DNA is blocked by overlapping CpG methylation. A single EcoR V site is found in the tetracycline resistance gene of pBR322.

Source: E.coli that carries the EcoR V gene from plasmid J62 pLG74

Reaction conditions

1X FastGene® Buffer III. 37°C 1X FastGene® FastCut Buffer, 37°C

FastGene® FastCut Buffer

FastGene® restriction enzyme can cut substrate DNA in 5-15 min with FastGene® FastCut Buffer.

1X FastGene® Buffer III

50 mM Tris-HCl (pH 7.9 at 25°C)

100 mM NaCl 10 mM MgCl₂ 100 µg/ml BSA

Unit definition

One unit is defined as the amount of enzyme required for complete digestion of 1 μg bacteriophage λ at 37°C for 1 hr in 50 µl reaction mixtures.

Quality control

- Unit definition assay Overdigestion assay
- Endonuclease assay
- Extreme pure assay

Standard reaction condition

- Normal protocol

Χ μΙ
5 μΙ
t 1 μl
up to 50 μl
ì

[→] Incubate at 37°C for 1 hr

- Fast protocol

p		
Component	Final Conc.	Volume
Substrate DNA	1 μg	Χ μΙ
10X FastGene® FastCut Buffer	1 X	5 μΙ
EcoR V	20 unit	1 μΙ
Sterile water		up to 50 μl
Insubate at 27°C for 15 min		

[→] Incubate at 37°C for 15 min

* We recommend 5-10 units of enzyme per μg DNA and 10-20 units for genomic DNA in a 1 h digest.