FastGene®

# Restriction Enzyme Sph I

II 37° 65°

| Cat.# | Size | Conc. |
|---|---|---|
| FG-SphI | 600 units | 5 units/µl |

**Store at -20℃**

**Supplied with:**
10X FastGene® Buffer II (FG-REB2)
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

**Source**: *Streptomyces phaeochromogenes*

## Reaction conditions
1X FastGene® Buffer II, 37℃
1X FastGene® FastCut Buffer, 37℃

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

## 1X FastGene® Buffer II
10 mM Tris-HCl (pH 7.9 at 25℃)
50 mM NaCl
10 mM $MgCl_2$
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℃ for 1 hr in 50 µl reaction mixtures.

## Quality control
- Unit definition assay
- Overdigestion assay
- Endonuclease assay
- Extreme pure assay

## Dilution buffer
FastGene® Diluent B

## Heat Inactivation
Sph I can be inactivated at 65℃ for 20 min.

## Methylation sensitivity
*dam* methylation: Not sensitive
*dcm* methylation: Not sensitive
CpG methylation: Not sensitive

## Prolonged incubation
A minimum amount of enzyme required to digest 1µg substrate DNA for 16 hr; 0.13 U.

## Relative activity in FastGene® Buffers

| | |
|---|---|
| FastGene® Buffer I: | 50% |
| FastGene® Buffer II: | 100% |
| FastGene® Buffer III: | 50% |
| FastGene® Buffer IV: | 75% |
| FastGene® FastCut Buffer: | 100% |

## Note
It produces a 3' CATG extension, which can be efficiently ligated to DNA fragments cleaved by Nla III. It is not affected by *dam*, *dcm*, or mammalian CpG methylation. Phenol extraction is not suitable to isolate Sph I-cleaved DNA fragments due to a tight association of Sph I with DNA. Low concentration of NaCl enhances aggregation.

## Standard reaction condition

- Normal protocol

| Component | Final Conc. | Volume |
|---|---|---|
| Substrate DNA | 1 µg | X µl |
| 10X FastGene® Buffer II | 1 X | 5 µl |
| Sph I | 5 unit | 1 µl |
| Sterile water | | up to 50 µl |

→ Incubate at 37℃ for 1 hr

- Fast protocol

| Component | Final Conc. | Volume |
|---|---|---|
| Substrate DNA | 1 µg | X µl |
| 10X FastGene® FastCut Buffer | 1 X | 5 µl |
| Sph I | 5 unit | 1 µl |
| Sterile water | | up to 50 µl |

→ Incubate at 37℃ 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.