

CERTIFICATE OF ANALYSIS

SgrDI

#ER2031 200 u

Lot: Expiry Date:

5'...**C G↓T C G A C G**...3'

3'...**G C A G C T↑G C**...5'

Concentration: 5 u/μl

Source: *E.coli* that carries the cloned *sgrDIR*
gene from *Streptomyces griseus* RFL6

Supplied with: 1 ml of 10X Buffer R
1 ml of 10X Buffer Tango™

Store at -20°C



In total 3 vials.

BSA included: Lot# BSA62-313P

RECOMMENDATIONS

1X Buffer R (for 100% SgrDI digestion)

10 mM Tris-HCl (pH 8.5), 10 mM MgCl₂, 100 mM KCl,
0.1 mg/ml BSA.

Incubation temperature

37°C.

Unit Definition

One unit is defined as the amount of SgrDI required to digest 1 μg of lambda DNA-Eco81I fragments in 1 hour at 37°C in 50 μl of recommended reaction buffer.

Dilution

Dilute with Dilution Buffer (#B19): 10 mM Tris-HCl (pH 7.4 at 25°C), 100 mM KCl, 1 mM EDTA, 1 mM DTT, 0.2 mg/ml BSA and 50% glycerol.

Double Digests

Tango™ Buffer is provided to simplify buffer selection for double digests. 98% of Fermentas restriction enzymes are active in a 1X or 2X concentration of Tango™ Buffer. Please refer to the Fermentas Catalog or go to www.fermentas.com/doubledigest to choose the best buffer for your experiments.

Storage Buffer

SgrDI is supplied in: 10 mM Tris-HCl (pH 7.5 at 25°C), 300 mM KCl, 0.1 mM EDTA, 1 mM DTT, 0.2 mg/ml BSA and 50% glycerol.

Recommended Protocol for Digestion

- Add:

nuclease-free water	16 μ l
10X Buffer R	2 μ l
DNA (0.5-1 μ g/ μ l)	1 μ l
SgrDI	0.5-2 μ l*
- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours*.

The digestion reaction may be scaled either up or down.

Recommended Protocol for Digestion of PCR Products Directly after Amplification

- Add:

PCR reaction mixture	10 μ l (~0.1-0.5 μ g of DNA)
nuclease-free water	18 μ l
10X Buffer R	2 μ l
SgrDI	1-2 μ l*
- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours*.

* See Star Activity.

Thermal Inactivation

SgrDI is inactivated by incubation at 65°C for 20 min.

ENZYME PROPERTIES

Enzyme Activity in Fermentas REase Buffers, %

B	G	O	R	Tango™	2X Tango™
0-20	0-20	0-20	100	NR	100

NR – buffer is not recommended, because of high star activity.

Star Activity

An excess of SgrDI (5 u/ μ g DNA x 16 hours) or low salt concentration may result in star activity.

Methylation Effects on Digestion

Dam: never overlaps – no effect.

Dcm: never overlaps – no effect.

CpG: completely overlaps – blocked.

EcoKI: never overlaps – no effect.

EcoBI: never overlaps – no effect.

Stability during Prolonged Incubation

A minimum of 0.3 units of the enzyme is required for complete digestion of 1 μ g of lambda DNA in 16 hours at 37°C.

Digestion of Agarose-embedded DNA

A minimum of 5 units of the enzyme is required for complete digestion of 1 μ g of agarose-embedded lambda DNA in 16 hours.

Compatible Ends

Sall, Eco88I, SmaI, XhoI.

Number of Recognition Sites in DNA

λ	Φ X174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
1	0	0	0	0	0	0

For **QUALITY CONTROL ASSAY DATA** see back page

QUALITY CONTROL ASSAY DATA

Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after a 40-fold overdigestion) with SgrDI (2.5 u/μg lambda DNA x 16 hours (see Star Activity).

Ligation/Recutting Assay

After a 30-fold overdigestion (2 u/μg DNA x 16 hours) with SgrDI, more than 80% of the digested DNA fragments can be ligated at a 5'-termini concentration of 0.02 μM. More than 95% of these sites can be recut.

Labeled Oligonucleotide (LO) Assay

No detectable degradation of single-stranded or double-stranded labeled oligonucleotides occurred during incubation with 10 units of SgrDI for 4 hours.

Blue/White Cloning Assay

A mixture of pUC57/HindIII, pUC57/Eco32I and pUC57/PstI digests was incubated with 10 units of SgrDI for 16 hours. After religation and transformation, the background level of white colonies was 0.4%.

Quality authorized by:



Jurgita Zilinskiene

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Please refer to www.fermentas.com for Material Safety Data Sheet of the product.