



CERTIFICATE OF ANALYSIS

TsoI

#ER1991 50 u

Lot: Expiry Date:

A

5'... T A G C C A (N)₁₁↓...3'

3'... A T C G G T (N)₉↑...5'

T

Concentration: 3 u/μl
Source: *Thermus scotoductus* RFL4
Supplied with: 1 ml of 10X Buffer G
1 ml of 10X Buffer Tango™
0.1 ml of 50X SAM (2.5 mM)

Store at -20°C



In total 4 vials.

BSA included: Lot# BSA62-313P

RECOMMENDATIONS

1X [Buffer G]+SAM (for 100% TsoI digestion)

[10 mM Tris-HCl (pH 7.5 at 37°C), 10 mM MgCl₂,
50 mM NaCl, 0.1 mg/ml BSA] +
0.05 mM S-adenosylmethionine (SAM).

Incubation Temperature

55°C*

Unit Definition

One unit is defined as the amount of TsoI at which no change in the fragmentation pattern is observed with further increase of enzyme. 1 μg of lambda DNA is incubated with the enzyme for 1 hour at 55°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.

1X Tango™ Buffer:

33 mM Tris-acetate (pH 7.9 at 37°C), 10 mM magnesium acetate, 66 mM potassium acetate, 0.1 mg/ml BSA.

*Incubation at 37°C results in 10% activity.

Storage Buffer

Tsol is supplied in: 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.

Recommended Protocol for Digestion

- Add:

nuclease-free water	16 µl
10X Buffer G	2 µl
DNA (0.5-1 µg/µl)	1 µl
50X SAM	0.4 µl
Tsol	0.5-2 µl**
- Mix gently and spin down for a few seconds.
- Incubate at 55°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 µl	(~0.1-0.5 µg of DNA)
nuclease-free water	18 µl	
10X Buffer G	2 µl	
50X SAM	0.6 µl	
Tsol	1-2 µl**	
- Mix gently and spin down for a few seconds.
- Incubate at 55°C for 1-16 hours**.

** See Note.

Thermal Inactivation

Tsol is inactivated by incubation at 80°C for 20 min.

ENZYME PROPERTIES

Enzyme Activity in Fermentas REase Buffers, %

B _{+SAM}	G _{+SAM}	O _{+SAM}	R _{+SAM}	Tango™ _{+SAM}	2X Tango™ _{+SAM}
NR	100	50-100	0-20	50-100	20-50

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

Methylation Effects on Digestion

- Dam: never overlaps – no effect.
- Dcm: may overlap – no effect.
- CpG: never overlaps – no effect.
- EcoKI: may overlap – effect not determined.
- EcoBI: never overlaps – no effect.

Stability during Prolonged Incubation

A minimum of 1 unit of the enzyme is required for digestion of 1 µg of lambda DNA in 16 hours at 37°C.

Number of Recognition Sites in DNA

λ	ΦX174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
43	6	4	1	1	3	4

Note

- A large excess of Tsol (7.5 u/µg DNA x 16 hours) may result in star activity.
- Tsol requires SAM for activity.
- Complete cleavage of some substrates by Tsol is difficult to achieve.
- Tsol may remain associated with the cleaved DNA. This may cause DNA band shifting during electrophoresis. To avoid an atypical DNA band pattern, use the 6X Loading Dye & SDS Solution (#R1151) for sample preparation or heat the digested DNA in the presence of SDS prior to electrophoresis.

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 96-fold overdigestion (6 u/μg lambda DNA x 16 hours) with TsoI.

Ligation/Recutting Assay

After a 10-fold overdigestion (0.6 u/μg DNA x 17 hours) with TsoI, approximately 80% of the digested DNA fragments can be ligated at a 5'-termini concentration of 0.18 μM. None of these sites can be recut due to methylation of the recognition sequence by TsoI.

Labeled Oligonucleotide (LO) Assay

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

Quality authorized by:



Jurgita Zilinskiene

PRODUCT USE LIMITATION.

This product is developed, designed and sold exclusively *for research purposes and in vitro use only*. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.

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