

#### **CERTIFICATE OF ANALYSIS**

## RsaI

**#ER1122** 5000 u

Lot: \_\_\_\_ Expiry Date: \_\_\_\_

5'...**G T↓A C**...3'

3'...**C A↑T G**...5'

Concentration: 10 u/µl

Source: Rhodopseudomonas sphaeroides

Supplied with: 2x1 ml of 10X Buffer Tango<sup>™</sup>

Store at -20°C















In total 3 vials.

BSA included



#### RECOMMENDATIONS

**1X Buffer Tango**<sup>™</sup> (for 100% Rsal digestion)

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

## **Incubation temperature**

37°C.

#### **Unit Definition**

One unit is defined as the amount of Rsal required to digest 1  $\mu$ g of lambda DNA in 1 hour at 37°C in 50  $\mu$ l 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<sup>™</sup> 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<sup>™</sup> Buffer. Please refer to the Fermentas Catalog or go to <a href="https://www.fermentas.com/doubledigest">www.fermentas.com/doubledigest</a> to choose the best buffer for your experiments.

## **Storage Buffer**

Rsal 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.

Rev. 7

## **Recommended Protocol for Digestion**

• Add:

nuclease-free water	16 µl
10X Buffer Tango <sup>™</sup>	2 µl
DNA (0.5-1 μg/μl)	1 µl
Rsal	0.5-2 µl

- 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  $\mu$ l (~0.1-0.5  $\mu$ g of DNA) nuclease-free water 18  $\mu$ l 10X Buffer Tango<sup>TM</sup> 2  $\mu$ l Rsal 1-2  $\mu$ l

- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours.

#### **Thermal Inactivation**

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

#### **ENZYME PROPERTIES**

## **Enzyme Activity in Fermentas REase Buffers, %**

В	G	0	R	Tango <sup>™</sup>	2X Tango <sup>™</sup>
50-100	20-50	0-20	0-20	100	0-20

## **Methylation Effects on Digestion**

Dam: never overlaps — no effect. Dcm: never overlaps — no effect.

CpG: may overlap – cleavage impaired.

EcoKI: never overlaps — no effect. EcoBI: never overlaps — no effect.

## **Stability during Prolonged Incubation**

A minimum of 0.2 units of the enzyme is required for complete digestion of 1  $\mu$ g of lambda DNA in 16 hours at 37°C.

## **Number of Recognition Sites in DNA**

_	λ	ФХ174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
	113	11	3	3	3	2	19

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 160-fold overdigestion with Rsal (10  $u/\mu g$  lambda DNA x 16 hours).

## **Ligation/Recutting Assay**

After a 50-fold overdigestion (3  $u/\mu g$  DNA x 17 hours) with Rsal, more than 95% of the digested DNA fragments can be ligated at a 5'-termini concentration of 1.0  $\mu$ M. More than 95% of these sites can be recut.

## **Labeled Oligonucleotide (LO) Assay**

No detectable degradation of single-stranded or doublestranded labeled oligonucleotides occurred during incubation with 10 units of Rsal 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 <a href="https://www.fermentas.com">www.fermentas.com</a> for Material Safety Data Sheet of the product.