

CERTIFICATE OF ANALYSIS

# Eam1104I (EarI)

#ER0232 1500 u

Lot: Expiry Date:

5'...**C T C T T C (N)**<sub>1</sub>↓...3'  
3'...**G A G A A G (N)**<sub>4</sub>↑...5'

Concentration: 10 u/μl  
Source: *E.coli* that carries the cloned  
*eam1104IR* gene from *Enterobacter*  
*amnigenus* RFL1104  
Supplied with: 1 ml of 10X Buffer Tango™

Store at -20°C



In total 2 vials.

BSA included: Lot# BSA62-313P

## RECOMMENDATIONS

**1X Buffer Tango™** (for 100% Eam1104I digestion)  
33 mM Tris-acetate (pH 7.9), 1 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 Eam1104I required  
to digest 1 μg of lambda DNA 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](http://www.fermentas.com/doubledigest) to choose the best  
buffer for your experiments.

### Storage Buffer

Eam1104I 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 $\mu$ l
10X Buffer Tango™	2 $\mu$ l
DNA (0.5-1 $\mu$ g/ $\mu$ l)	1 $\mu$ l
Eam1104I	0.5-2 $\mu$ 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 $\mu$ l (~0.1-0.5 $\mu$ g of DNA)
nuclease-free water	18 $\mu$ l
10X Buffer Tango™	2 $\mu$ l
Eam1104I	1-2 $\mu$ l
- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours.

## Thermal Inactivation

Eam1104I 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™
50-100	50-100	0-20	0-20	100	0-20

### Methylation Effects on Digestion

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

### Stability during Prolonged Incubation

A minimum of 0.5 units of the enzyme is required for complete digestion of 1  $\mu$ 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  $\mu$ g of agarose-embedded lambda DNA in 16 hours.

### Number of Recognition Sites in DNA

$\lambda$	$\Phi$ X174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
34	2	2	3	3	3	2

### Note

Particular sites in  $\lambda$  and plasmids DNA are difficult to cleave with Eam1104I, as well as with its prototype Ksp632I.

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 320-fold overdigestion with Eam1104I (20 u/μg lambda DNA x 16 hours).

## Ligation/Recutting Assay

After a 50-fold overdigestion (3 u/μg DNA x 17 hours) with Eam1104I, more than 95% of the digested DNA fragments can be ligated at a 5'-termini concentration of 0.2 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 Eam1104I 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](http://www.fermentas.com) for Material Safety Data Sheet of the product.