

PRODUCT INFORMATION

HinfI

#ER0801 2000 U

Lot: ___ Expiry Date: _

5'...**G**↓**A N T C**...3'

3'... **C T N A**↑**G**...5'

Concentration: 10 U/µL

Source: Haemophilus influenzae Rf

Supplied with: 1 mL of 10X Buffer R

1 mL of 10X Buffer Tango

Store at -20°C











In total 3 vials.

BSA included

RECOMMENDATIONS

1X Buffer R (for 100% Hinfl 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 Hinfl required to digest 1 μ g lambda DNA in 1 hour at 37°C in 50 μ 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

Thermo Scientific Tango Buffer is provided to simplify buffer selection for double digests. 98% of Thermo Scientific restriction enzymes are active in a 1X or 2X concentration of Tango™ Buffer. Please refer to www.thermoscientific.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.

Storage Buffer

Hinfl is supplied in: 10 mM Tris-HCl (pH 7.4 at 25°C), 100 mM KCl, 1 mM DTT, 1 mM EDTA, 0.2 mg/mL BSA and 50% glycerol.

Recommended Protocol for Digestion

• Add:

nuclease-free water $16 \mu L$ 10 X Buffer R $2 \mu L$ $DNA (0.5-1 \mu g/\mu L)$ $1 \mu L$ Hinfl $0.5-2 \mu L*$

- 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 μ L (~0.1-0.5 μ g of DNA) nuclease-free water 18 μ L 10X Buffer R 2 μ L Hinfl 1-2 μ L*

- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours.
- * This volume of the enzyme is recommended for preparations of standard concentrations (10 U/ μ L), whereas HC enzymes (50 U/ μ L) should be diluted with the Dilution Buffer to obtain 10 U/ μ L concentration.

Thermal Inactivation

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

ENZYME PROPERTIES

Enzyme Activity in Thermo Scientific REase Buffers, %

_	В	G	0	R	Tango	2X Tango
	0-20	20-50	50-100	100	50-100	50-100

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: may overlap — blocked.

Stability during Prolonged Incubation

A minimum of 0.1 units of the enzyme is required for complete digestion of 1 μ g of lambda DNA in 16 hours at 37°C.

Compatible Ends

 $G \downarrow A(A/T)TC - Pfel.$

Number of Recognition Sites in DNA

_	λ	ФХ174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
	148	21	10	5	6	9	27

For **CERTIFICATE OF ANALYSIS** see back page

CERTIFICATE OF ANALYSIS

Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after a 160-fold overdigestion with Hinfl (10 U/µg lambda DNA x 16 hours).

Ligation and Recleavage (L/R) Assay

The ligation and recleavage assay was replaced with LO test after validating experiments showed LO test ability to trace nuclease and phosphatase activities with sensitivity that is higher than L/R by a factor of 100.

Labeled Oligonucleotide (LO) Assay

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

Quality authorized by:



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.thermoscientific.com/onebio for Material Safety Data Sheet of the product.

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