

PRODUCT INFORMATION

MboI

#ER0811 300 U

Lot: ___ Expiry Date: _

5'....**↓G A T C**3'

3'... **C T A G**↑...5'

Concentration: 10 U/µL

Supplied with: 1 mL of 10X Buffer R

1 mL of 10X Buffer Tango

Store at -20°C

R 37º Dan





BSA included

www.thermoscientific.com/onebio

RECOMMENDATIONS

1X Buffer R (for 100% Mbol digestion) 10 mM Tris-HCl (pH 8.5), 10 mM MgCl₂, 100 mM KCl, 0.1 mg/mL BSA.

Incubation

37°C.

Unit Definition

One unit is defined as the amount of Mbol required to digest 1 μ g lambda DNA dam^- 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

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

Recommended Protocol for Digestion

• Add:

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

- 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 Mbol 1-2 μ L

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

Thermal Inactivation

Mbol 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
50-100	50-100	50-100	100	50-100	100

Methylation Effects on Digestion

Dam: completely overlaps – blocked.

Dcm: never overlaps – no effect. CpG: may overlap – no effect.

 $\label{eq:cokl:never} \mbox{EcoKI: never overlaps} - \mbox{no effect}.$

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

BamHI, BcII, BgIII, Psul

Number of Recognition Sites in DNA

λ	ФХ174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
116	0	22	15	15	15	7

Note

- Mbol is blocked by overlapping dam methylation. To avoid dam methylation, use a dam⁻, dcm⁻ strain such as GM2163 (#M0099).
- Mbol, Bsp143I and DpnI all recognize the same sequence but have different methylation sensitivities and cleavage sites.

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 MboI (10 U/µg lambda *dam*⁻ 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 Mbol 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.thermoscientific.com/onebio for Material Safety Data Sheet of the product.

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