

PRODUCT INFORMATION

MboII

#ER0822 1500 U

Lot: ____ **Expiry Date:** __

5'... **G A A G A (N)**₈↓...3'

3'... **C T T C T (N)**₇↑...5'

Concentration: 5 U/μL

Source: *E.coli* that carries the cloned *mbollR* gene from *Moraxella bovis*

Supplied with: 1 mL of 10X Buffer B
1 mL of 10X Buffer Tango

Store at -20°C



BSA included

www.thermoscientific.com/onebio

RECOMMENDATIONS

1X Buffer B (for 100% MboII digestion)

10 mM Tris-HCl (pH 7.5), 10 mM MgCl₂, 0.1 mg/mL BSA.

Incubation temperature

37°C.

Unit Definition

One unit is defined as the amount of MboII required to digest 1 μg of lambda DNA *dam*⁻ 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

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

Mboll 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 μ L
10X Buffer B	2 μ L
DNA (0.5-1 μ g/ μ L)	1 μ L
Mboll	0.5-2 μ 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 B	2 μ L
Mboll	1-2 μ L*
- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours*.

* See Star Activity.

Thermal Inactivation

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

ENZYME PROPERTIES

Enzyme Activity in Thermo Scientific REase Buffers, %

B	G	O	R	Tango	2X Tango
100	50-100	20-50	0-20	50-100	20-50

Star Activity

An excess of MbolI (15 U/ μ g DNA x 1 hour) may result in star activity.

Methylation Effects on Digestion

Dam: may overlap – blocked.

Dcm: never overlaps – no effect.

CpG: may overlap – no effect.

EcoKI: never overlaps – no effect.

EcoBI: may overlap – no effect.

Stability during Prolonged Incubation

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

Number of Recognition Sites in DNA

λ	Φ X174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
130	11	11	8	7/8	9	11

For **CERTIFICATE OF ANALYSIS** see back page

Note

- MbolI is blocked by overlapping *dam* methylation. To avoid *dam* methylation, use a *dam*⁻, *dcm*⁻ strain such as GM2163 (#M0099).
- MbolI produces DNA fragments that have a single-base 3'-extension which are more difficult to ligate than blunt-ended fragments.
- MbolI may remain associated with the cleaved DNA. This may cause DNA band shifting during electrophoresis. To avoid atypical DNA band patterns, use the 6X DNA Loading Dye&SDS Solution (#R1151) for sample preparation or heat the digested DNA in the presence of SDS prior to electrophoresis.

CERTIFICATE OF ANALYSIS

Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after a 10-fold overdigestion with MbolI (10 U/μg lambda DNA *dam*⁻ x 1 hour) (*see* Star Activity).

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 double-stranded labeled oligonucleotides occurred during incubation with 10 units of MbolI 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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