

PRODUCT INFORMATION

BglII

#ER0081 500 U

Lot: _____ Expiry Date: _

5'...A↓G A T C T...3'
3'...T C T A G↑A...5'

Concentration: 10 U/μL
Supplied with: 1 mL of 10X Buffer 0
 1 mL of 10X Buffer Tango

Store at -20°C



RECOMMENDATIONS

1X Buffer 0 (for 100% BglII digestion)

50 mM Tris-HCl (pH 7.5), 10 mM MgCl₂, 100 mM NaCl,
0.1 mg/μL BSA.

Incubation temperature

37°C.

Unit Definition

One unit is defined as the amount of BglII 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/μL 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/μL BSA.

Storage Buffer

BgIII is supplied in: 10 mM Tris-HCl (pH 8.2 at 25°C), 200 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.5 mg/μL BSA and 50% glycerol.

Recommended Protocol for Digestion

- Add:

nuclease-free water	16 μL
10X Buffer O	2 μL
DNA (0.5-1 μg/μL)	1 μL
BgIII	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 μL (~0.1-0.5 μg of DNA)
nuclease-free water	18 μL
10X Buffer O	2 μL
BgIII	1-2 μL
- Mix gently and spin down for a few seconds.
- Incubate at 37°C for 1-16 hours.

Thermal Inactivation

BgIII is not inactivated by incubation at 80°C for 20 min.

Inactivation Procedure

- To prepare the digested DNA for electrophoresis:
 - stop the digestion reaction by adding 0.5 M EDTA, pH 8.0 (#R1021), to achieve a 20 mM final concentration. Mix thoroughly, add an electrophoresis loading dye and load onto gel.
- To prepare DNA suitable for further enzymatic reactions:
 - extract with phenol/chloroform, precipitate with ethanol or isopropanol, wash the pellet with 75% cold ethanol and air-dry;
 - dissolve DNA in either nuclease-free water, TE buffer, or a buffer suitable for further applications;
 - check the DNA concentration in the solution.

For **ENZYME PROPERTIES** and **CERTIFICATE OF ANALYSIS**

see back page

ENZYME PROPERTIES

Enzyme Activity in Thermo Scientific REase Buffers, %

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

Methylation Effects on Digestion

Dam: completely overlaps— no effect.

Dcm: never overlaps – no effect.

CpG: never overlaps – no effect.

EcoKI: never overlaps – no effect.

EcoBI: may overlap – cleavage impaired.

Stability during Prolonged Incubation

A minimum of 0.1 units of the enzyme is required for complete digestion of 1 µg 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 µg of agarose-embedded lambda DNA in 16 hours.

Compatible Ends

BamHI, BclI, Bsp143I, MboI, PstI

Number of Recognition Sites in DNA

λ	ΦX174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
6	0	0	0	0	0	1

CERTIFICATE OF ANALYSIS

Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after a 160-fold overdigestion with BglII (10 U/µg lambda DNA × 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 double-stranded labeled oligonucleotides occurred during incubation with 10 units of BglII for 4 hours.

Blue/White (B/W) Cloning Assay

The B/W assay was replaced with LO test after validating experiments showed LO test ability to detect nuclease and phosphatase activities with sensitivity that equals to that of B/W test.

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.

© 2012 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific Inc. and its subsidiaries.