Thermo SCIENTIFIC

PRODUCT INFORMATION

BveI (BspMI)

#ER1741 250 U

Lot: ___ **Expiry Date: _**

5'...**A C C T G C(N)$_4$**↓...3'
3'...**T G G A C G(N)$_8$**↑...5'

Concentration:	5 U/µL
Source:	*E.coli* that carries the cloned *bvelR* gene from *Brevundimonas vesicularis* RFL1
Supplied with:	1 mL of 10X Buffer O 1 mL of 10X Buffer Tango 2 x 25 µL of 50X oligonucleotide (0.025 mM)

Store at -20°C

BSA included

www.thermoscientific.com/onebio

RECOMMENDATIONS

[1X Buffer O] + oligonucleotide* (for 100% BveI digestion)
[50 mM Tris-HCl (pH 7.5), 10 mM $MgCl_2$, 100 mM NaCl, 0.1 mg/mL BSA] + 0.5 µM of oligonucleotide.

Incubation temperature
37°C.

Unit Definition
One unit is defined as the amount of BveI at which no change in the fragmentation pattern is observed with further increase of the enzyme. 1 µg of lambda DNA is incubated with BveI for 1 hour at 37°C in 50 µL of recommended reaction buffer. The cleavage of DNA by BveI is never complete.

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.

* Plasmid DNAs containing single BveI sites are resistant to cleavage. Digestion of slowly cleaved and resistant sites can be significantly enhanced by the addition of a 0.5µM oligonucleotide containing BveI recognition sequence.

Storage Buffer

BveI is supplied in: 10 mM Tris-HCl (pH 7.5 at 25°C), 150 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 µL
10X Buffer O	2 µL
DNA (0.5-1 µg/µL)	1 µL
50X oligonucleotide (0.025 mM)	0.4 µL
BveI	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
50X oligonucleotide (0.025 mM)	0.6 µL
BveI	1-2 µL**

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

** *See* Overdigestion Assay.

Thermal Inactivation

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

ENZYME PROPERTIES

Enzyme Activity in Thermo Scientific REase Buffers, %

B_{+oligo}	G_{+oligo}	O_{+oligo}	R_{+oligo}	$Tango_{oligo}$	$2X\ Tango_{+oligo}$
0-20	20-50	**100**	20-50	50-100	100

Methylation Effects on Digestion

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

Stability during Prolonged Incubation

A minimum of 0.2 units of the enzyme is required for digestion of 1 µg of lambda DNA in 16 hours at 37°C.

Number of Recognition Sites in DNA

λ	ΦX174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
41	3	1	0	1	1	3

Note

- At least two copies of BveI recognition site are required for efficient cleavage.
- BveI 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.

For **CERTIFICATE OF ANALYSIS** *see* back page

CERTIFICATE OF ANALYSIS

Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after an 80-fold overdigestion with BveI (5 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 double-stranded labeled oligonucleotides occurred during incubation with 10 units of BveI 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.