Thermo SCIENTIFIC

PRODUCT INFORMATION

BseXI (BbvI)

#ER1452 500 U

Lot: ___ **Expiry Date: _**

5'...**G C A G C(N)$_8$** ↓...3'
3'...**C G T C G(N)$_{12}$**↑...5'

Concentration: 3 U/µL
Supplied with: 1 mL of 10X Buffer BseXI

Store at -20°C

BSA included

www.thermoscientific.com/onebio

RECOMMENDATIONS

1X Buffer BseXI (for 100% BseXI digestion)
50 mM Tris-HCl (pH 7.5), 2 mM $MgCl_2$, 100 mM NaCl, 0.1 mg/mL BSA.

Incubation temperature
65°C*****.

Unit Definition
One unit is defined as the amount of BseXI required to digest 1 µg of pBR322 DNA in 1 hour at 65°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
Please go to www.thermoscientific.com/doubledigest to choose the best buffer for your experiments.

Storage Buffer
BseXI is supplied in: 10 mM Tris-HCl (pH 7.4 at 25°C), 200 mM NaCl, 1 mM DTT, 1 mM EDTA, 0.2 mg/mL BSA and 50% glycerol.

***** Incubation at 37°C results in 10% activity.

Recommended Protocol for Digestion

- Add:

nuclease-free water	16 µL
10X Buffer BseXI	2 µL
DNA (0.5-1 µg/µL)	1 µL
BseXI	0.5-2 µL**

- Mix gently and spin down for a few seconds.
- Incubate at 65°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 BseXI	2 µL
BseXI	1-2 µL**

- Mix gently and spin down for a few seconds.
- Incubate at 65°C for 1-16 hours**.

** *See* Star Activity.

Thermal Inactivation

BseXI is inactivated by incubation at 80°C for 20 min.

ENZYME PROPERTIES

Enzyme Activity in Thermo Scientific REase Buffers, %

Buffer BseXI – 100%.
None of the standard buffers is recommended, because of high star activity.

Star Activity

A large excess of the enzyme (3 U/µg DNA x 16 hours) may result in star activity.

Methylation Effects on Digestion

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

Stability during Prolonged Incubation

A minimum of 0.3 units of the enzyme is required for complete digestion of 1 µg of pBR322 in 16 hours at 65°C.

Number of Recognition Sites in DNA

λ	ΦX174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
199	14	21	12	12	12	10

Note

BseXI 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 a 32-fold overdigestion with BseXI (2 U/µg pBR322 DNA x 16 hours) (*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 5 units of BseXI 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.