



## PRODUCT INFORMATION

# BspPI (AlwI)

#ER1322      500 U

Lot: \_\_\_      Expiry Date: \_\_\_

5'...G G A T C (N)<sub>4</sub>↓...3'  
3'...C C T A G (N)<sub>5</sub>↑...5'

Concentration:      2 U/μL  
Supplied with:      1 mL of 10X Buffer Tango

**Store at -20°C**



BSA included

[www.thermoscientific.com/onebio](http://www.thermoscientific.com/onebio)

## RECOMMENDATIONS

**1X Thermo Scientific Tango Buffer** (for 100% BspPI digestion)

33 mM Tris-acetate (pH 7.9), 10 mM magnesium acetate, 66 mM potassium acetate, 0.1 mg/mL BSA.

### Incubation temperature

55°C\*.

### Unit Definition

One unit is defined as the amount of BspPI required to digest 1 μg of lambda DNA *dam*<sup>-</sup> in 1 hour at 55°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

Tango™ Buffer provided simplifies buffer selection for double digests. 98% of Thermo Scientific restriction enzymes are active in a 1X or 2X concentration of Tango Buffer. Please go to [www.thermoscientific.com/doubledigest](http://www.thermoscientific.com/doubledigest) to choose the best buffer for your experiments.

### Storage Buffer

BspPI is supplied in: 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.

\* Incubation at 37°C results in 30% activity.

## Recommended Protocol for Digestion

- Add:

nuclease-free water	16 $\mu$ L
10X Buffer Tango	2 $\mu$ L
DNA (0.5-1 $\mu$ g/ $\mu$ L)	1 $\mu$ L
BspPI	0.5-2 $\mu$ L
- Mix gently and spin down for a few seconds.
- Incubate at 55°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 $\mu$ L (~0.1-0.5 $\mu$ g of DNA)
nuclease-free water	18 $\mu$ L
10X Buffer Tango	2 $\mu$ L
BspPI	1-2 $\mu$ L
- Mix gently and spin down for a few seconds.
- Incubate at 55°C for 1-16 hours.

## Thermal Inactivation

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

## ENZYME PROPERTIES

### Enzyme Activity in Thermo Scientific REase Buffers, %

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

### Methylation Effects on Digestion

Dam: completely overlaps – blocked.

Dcm: may overlap – no effect.

CpG: may overlap – no effect.

EcoKI: never overlaps – no effect.

EcoBI: may overlap – no effect.

### Stability during Prolonged Incubation

A minimum of 0.5 units of enzyme is required for complete digestion of 1  $\mu$ g of lambda DNA in 16 hours at 55°C.

### Number of Recognition Sites in DNA

$\lambda$	$\Phi$ X174	pBR322	pUC57	pUC18/19	pTZ19R/U	M13mp18/19
58	0	12	10	10	10	4

### Note

BspPI is blocked by overlapping *dam* methylation. To avoid *dam* methylation, use a *dam*<sup>-</sup>, *dcm*<sup>-</sup> strain such as GM2163 (#M0099).

For **CERTIFICATE OF ANALYSIS** see back page

# CERTIFICATE OF ANALYSIS

## Overdigestion Assay

No detectable change in the specific fragmentation pattern is observed after a 80-fold overdigestion with BspPI (5 U/ $\mu$ g lambda DNA *dam*<sup>-</sup> 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 5 units of BspPI 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](http://www.thermoscientific.com/onebio) for Material Safety Data Sheet of the product.

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