

Nt.BspQI

Product Number: RE05882

Shipping and Storage

-20°C.

Components

| Component | Specifications |
|--------------------|----------------|
| Nt.BspQI (10 U/μL) | 200μL |
| 10×Cut Buffer C | 2×1mL |

Description

Nt.BspQI is a type of endonuclease that cleaves only one strand of dsDNA substrate; Create incisions on the dsDNA substrate without incising the dsDNA.

Suggested reaction conditions

1. 10×Cut Buffer C.
2. Incubate at 50°C.
3. Prepare the reaction system according to the "DNA rapid enzyme digestion process".
4. This product has 100% activity when subjected to enzymatic digestion at 37°C.

Inactivation conditions

Incubate at 80°C for 20 minutes.

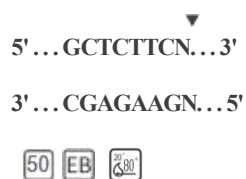
Definition of Activity




1 active unit (U) refers to the amount of enzyme required to completely convert 1μg of supercoiled pUC19 DNA into an open-loop form within 1 hour at 50°C in a 50μL reaction system.

Quality control

1. Ultra long incubation test: incubate 10 U Nt. BspQI and super spiral pUC19 DNA substrate at 50°C for 16 h, and detect no change in open loop DNA by agarose gel electrophoresis.
2. RNase residue detection: incubate 10 U Nt. BspQI and 500ng RNA at 37°C for 1 h, and use agarose gel electrophoresis to detect that more than 90% of RNA remains intact.

Icon annotation



1.  The optimal reaction temperature is 50°C.
2.  For DNA methylated by EcoBI, splicing may be hindered
3.  Inactivation condition: Incubate at 80°C for 20 minutes.

Protocol

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1. DNA rapid enzymatic digestion process
1.1. Prepare the reaction system on ice according to the recommended sample addition sequence as follows:

| | |
|--------------------|------------|
| ddH ₂ O | up to 50μL |
| 10×Cut Buffer C | 5μL |
| Substrate DNA | 1μg |
| Nt.BspQI (10 U/μL) | 1μL |
| Total | 20μL |

The DNA substrate should not contain phenol, chloroform, ethanol EDTA, Detergent or high concentration salt, otherwise it will affect the activity of Nt.BspQI enzyme;

- 1.2. Gently suck or tap the tube wall to mix well (without vortexing), then centrifuge instantly to collect hanging wall droplets;
- 1.3. Incubate at 50°C for 30 minutes to 1 hour;
- 1.4. Incubate at 80°C for 20 minutes to inactivate the enzyme, stop the reaction, or terminate the reaction through adsorption column or phenol/chloroform purification.

2. Note

- 2.1. The volume of enzyme added to the reaction system should not exceed 10% of the total volume to avoid excessive glycerol in the enzyme causing star activity;
- 2.2. The additives (such as glycerol, salt) in the storage buffer for restriction endonucleases are the same as the pollutants (such as salt, EDTA, or ethanol) in the substrate solution. The smaller the reaction volume, the stronger the inhibitory effect on the enzyme cleavage reaction.

The number of enzyme cleavage sites in different DNA

| λDNA | ΦX174 | pBR322 | pUC57 | pUC18/19 | SV40 | M13mp18/19 | Adeno2 |
|------|-------|--------|-------|----------|------|------------|--------|
| 10 | 1 | 1 | 1 | 1 | 0 | 0 | 7 |

The influence of methylation modification

| Dam | Dcm | CpG | EcoKI | EcoBI |
|-----------|-----------|-----------|-----------|--------------|
| no effect | no effect | no effect | no effect | Shear effect |