

## Tinzyme Co., Limited

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# **Cas9** Nuclease

### **Product Number: CAS09**

### **Shipping and Storage**

Store at  $-30 \sim -15^{\circ}$ C and transport at  $\leq 0^{\circ}$ C.

### Components

Component	CAS09	CAS09
	500pmol	1000pmol
Cas9 Nuclease (1µM)	50µl	250µl
Cas9 Nuclease Reaction Buffer (10×)	1ml	1ml

## Description

Cas9 Nuclease is an RNA directed sequence specific double stranded DNA endonuclease.Wizard RNA identifies target sites by complementing with the target sequence and carries Cas9 Nuclease onto the target DNA.Cas9 Nuclease has two cleaving active centers, each cutting two strands of target DNA, resulting in DNA double strand breaks.The cleavage site is located within the target region and three bases away from NGG (PAM).

#### Source

This product is a high-purity Cas9 active protein expressed and purified in Escherichia coli using the Cas9 gene cloned from Streptococcus pyogenes.

### Application

Double stranded DNA cleavage; Genomic modification.

### Unit definition

1 unit refers to the amount of enzyme required to add 0.5pmol of dNTP to acid insoluble precipitate during a 10 minute reaction at 30°C.

#### Protocol

1. Prepare the system reaction solution according to the following suggestions:

Reagent	Volume
Nuclease-free ddH <sub>2</sub> O	20µ1
Cas9 Nuclease Reaction Buffer (10×)	3µ1
sgRNA	3µl
Cas9 Nuclease	1µl
Total	27µl

Note:1)To ensure the highest cutting efficiency, the molar ratio of Cas9 Nuclease and sgRNA to target DNA should be at least 10:10:1 or higher.

2)Generally, use  $30\mu$ l system , but it can also be scaled up in equal proportions.

3)Please dilute sgRNA to 300nM using Nuclease free  $ddH_2O$  before the experiment.

4)Please wear a mask and use Nuclease free consumables and reagents to avoid the degradation of sgRNA during the experiment.

2. Incubate at 37°C for 10 minutes.

3. Add 3µl of 30nM DNA.

### For Research Use Only



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Note: Dilute DNA to 30nM using Nuclease free ddH<sub>2</sub>O.

- 4. Shake well and centrifuge briefly for collection.
- 5. Incubate at 37 °C for 1 hour.
- The reaction products can be directly analyzed by agarose gel electrophoresis.
  Note: If electrophoresis is not done immediately, EDTA can be added to terminate the reaction.

#### Note

This product is for scientific research purposes only and shall not be used for clinical medical diagnosis or other unreasonable purposes.