

AapCas12b Nuclease Instruction Manual

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|-------------------------|--------------------|-----------------------|---------|
| Product Number | AapCas12b Nuclease | Product Number | EDE0006 |
| Molecular Weight | 130.5 KDa | Form | Liquid |

I. Product Description

AapCas12b nuclease (also known as C2c1) is derived from *Alicyclobacillus acidophilus*. It is a crRNA-mediated nuclease that recognizes and cleaves double-stranded DNA (dsDNA) in the presence of a PAM (Protospacer Adjacent Motif) sequence (TTN). For single-stranded DNA (ssDNA) cleavage, the PAM sequence is not required. Upon binding to complementary ssDNA or dsDNA, the trans-cleavage activity of AapCas12b on non-specific ssDNA is activated. AapCas12b has an optimal reaction temperature of 60°C, making it more heat-resistant compared to Cas13a and Cas12a. This property makes it highly suitable for field nucleic acid detection applications when combined with loop-mediated isothermal amplification (LAMP).

II. Product Information

Product Components

| Component | EDE0006-50 | EDE0006-100 | EDE0006-500 |
|------------------------------------|------------------------|--------------------------|---------------------------|
| AapCas12b Nuclease | 5 μM*10 μL (50pmol) | 5 μM*20 μL (100 pmol) | 5 μM*100 μL (500 pmol) |
| AapCas12b Cleavage Buffer (10×) | 40 μL*1 tube | 80 μL*1 tube | 400 μL*1 tube |

Storage Conditions and Shelf Life

The product is stable for 1 year when stored at -20°C. **For long-term storage, it is recommended to store at -80°C. It is advised to aliquot the product based on the frequency of use to avoid repeated freeze-thaw cycles.**

Product Features

The product is prepared using a one-step purification process, retaining maximum enzymatic activity. It has been tested to show significantly higher activity compared to similar products.

▪ Activity Definition

In a 20 μ L reaction system at 60°C, the amount of Cas12b enzyme required to cleave 1 pmol of ssDNA probe within 1 minute defines 1 transU. For example, if a batch of AapCas12b enzyme exhibits trans-cleavage activity of 30 transU/pmol, it indicates that 1 pmol of this batch of AapCas12b enzyme can cleave 30 pmol of ssDNA probe under the specified reaction conditions within 1 minute.

▪ Quality Assurance

Sample Purity: ~95% (verified by SDS-PAGE).

III. Detection Steps

Required Reagents:

1. ssDNA reporter probe (labeled with FAM at the 5' end and BHQ1 at the 3' end).

Note: You can use the ssDNA reporter from our company or design your own.

| Product Name | Product Number |
|----------------------------|----------------|
| ssDNA reporter (DNA probe) | EDN-TD01 |

2. crRNA/gRNA: Forms a functional complex with Cas12b, which is specifically activated by the target sequence.

Note: Our company offers crRNA synthesis or you can custom-design crRNA.

| Name | Specification | Product Number |
|--------------------|---------------|----------------|
| sgRNA Biosynthesis | 2OD | EDN-RS01 |

For free design consultation, please contact: info@editxor.com

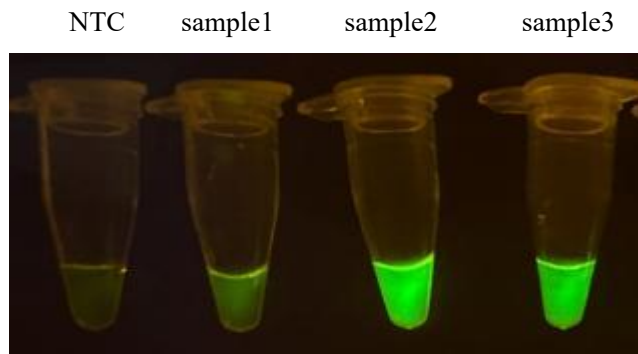
3. Isothermal Amplification Reaction Kit

Reaction System

| Component | Final Concentration | Volume (μ L) |
|------------------------------|---------------------|----------------------|
| 10 \times Cleavage Buffer | 1 \times | 2 μ L |
| 5 μ M AapCas12b Nuclease | 150-250 nM | 0.8 μ L (200 nM) |
| 500 nM crRNA | 150-250 nM | 8 μ L (200 nM) |
| 250 nM ssDNA Reporter | 150-250 nM | 1 μ L (200 nM) |
| 1 μ M DNA target | 150-250 nM | 4 μ L (200 nM) |
| DEPC H ₂ O | | |
| Total | 20 μ L | 20 μ L |

Reaction Conditions:

Use a real-time fluorescence quantitative PCR machine or an isothermal amplification instrument to detect the fluorescence signal. The reaction should occur at 60°C, and the fluorescence signal should be collected every 30 seconds. Alternatively, the fluorescence signal can be directly observed under a UV light.

Photograph Example**Precautions**

1. To prevent contamination by RNase, please keep the experimental area clean and tidy. Wear clean gloves and masks during operations. All consumables, such as pipette tips and centrifuge tubes, should be RNase-free.
2. Cas12 enzymes are prone to inactivation; store the enzyme at -20°C immediately after use.

Publishing Requirements

When using this product in publications, please acknowledge our company: Guangzhou Editgene Co. Ltd, China. Or EDITGENE CO.LTD if used within U.S. or Europe territory.