

# LbCas12a Nuclease Instruction Manual

Product Number	LbCas12a Nuclease	Product Number	EDE0005
Molecular Weight	146.8 KDa	Form	Liquid

## I. Product Description

LbCas12a nuclease (also known as Cpf1) is derived from Lachnospiraceae bacterium ND2006 strain. LbCas12a belongs to class 2 type V CRISPR effector proteins, and is a crRNA-mediated nuclease that recognizes and cleaves double-stranded DNA (dsDNA) in the presence of a PAM (Protospacer Adjacent Motif) sequence (TTTV). For single-stranded DNA (ssDNA) cleavage, the PAM sequence is not required. Upon binding to complementary ssDNA or dsDNA, the trans-cleavage activity of LbCas12a on non-specific ssDNA is activated. By designing single-stranded Reporter DNA labeled with fluorescent moieties or other small molecules at both ends, the detection and signal amplification of the DNA template by Cas12a can be realized. The results can be observed by fluorometer and test strips.

# **II. Product Information**

## **·Product Components**

Component	EDE0005-100	EDE0005-1000	EDE0005-2000
LbCas12a Nuclease	1 μM*100 μL (100pmol)	1 μM*500 μL *2 tube (1000 pmol)	1 μM*500 μL *4 tube (2000 pmol)
LbCas12a Cleavage Buffer (10×)	500 μL*1 tube	1mL*4 tube	1mL*7 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 30 µL reaction system at 37°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 LbCas12a enzyme exhibits trans-cleavage activity of 30 transU/pmol, it indicates that 1 pmol of this batch of LbCas12a 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	<b>Product Number</b>
crRNA Chemical Synthesis	20D	EDN-RH01

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

#### 3. Isothermal Amplification Reaction Kit / RPA Amplification Reaction Kit

Component	<b>Final Concentration</b>	Volume ( µL)
10× Cleavage Buffer	1×	3
1 µM LbCas12a Nuclease	33 nM	1
500 nM crRNA	33 nM	2
$2 \mu M$ ssDNA Reporter	400 nM	6
100 nM DNA target	3.3 nM	0.1
DEPC H <sub>2</sub> O		- CT
Total	30 µL	30 µL

#### **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 37°C, and the fluorescence signal should be collected every 30 seconds. Alternatively, the fluorescence signal can be directly observed under a UV light.

**Results:** 

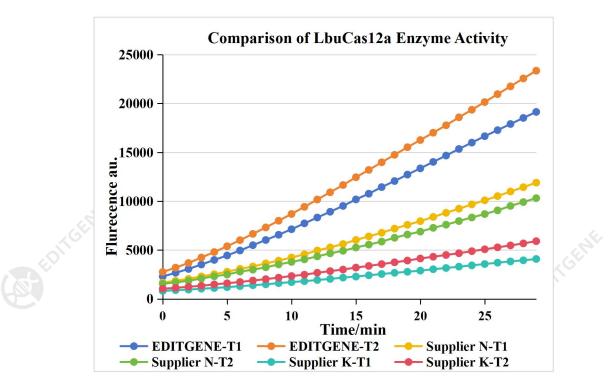


Fig 1. Results of Cas12a collateral cleavage at different company product.

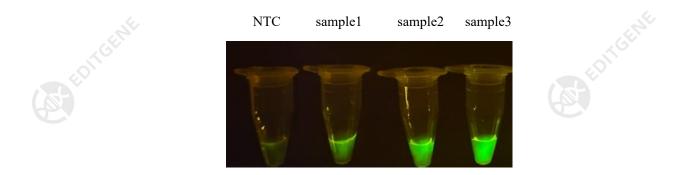
The horizontal coordinate refers to reaction time, and the vertical coordinate refers to detection result of Bio-rad CFX96 fluorescence PCR instrument. As shown in the graph, under the same condition, the cleaving efficiency of LbCas12a Nuclease is significantly higher than that from other suppliers.

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#### **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.

