

## Tinzyme Co., Limited

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# LwaCas13a (C2c2)

## Product Number: CAS13A

## **Shipping and Storage**

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

## Components

Component	CAS13A	CAS13A
	100pmol	1000pmol
LwaCas13a (10µM)	10µ1	100µl
LwaCas13a Reaction Buffer (10×)	1ml	2ml

## Description

LwaCas13a nuclease (C2c2) is an RNA endonuclease mediated by crRNA, originating from the Leptotrichia wadei strain. In the presence of PFS sequences in the target single stranded RNA, it can specifically recognize and cleave the target RNA.In addition, Cas13a also has trans cleavage activity (i.e., bypass cleavage activity/accessory cleavage activity), which means that when LwaCas13a protein binds to crRNA and target RNA to form a ternary complex, it is activated for trans cleavage activity against non-specific sequence single stranded RNA, shredding any sequence single stranded RNA in the system. This activity has also been used in the development of rapid detection kits for target nucleic acids.

#### Source

Obtained through E. coli recombination, expression, and purification, the expressed gene is derived from Leptotrichia wadei.

## Application

Cut any sequence of single stranded RNA, etc.

#### Unit definition

1 unit refers to the amount of Cas13a enzyme required to cleave 1 pmol ssRNA probe within 1 minute under 37°C reaction conditions

## Protocol

1. Prepare the system reaction solution (reverse shear experiment) according to the following suggestions:

Reagent	Volume	
LwaCas13a Reaction Buffer (10×)	2µ1	
LwaCas13a Nuclease (10µM)	0.05~0.5µl	
crRNA (10µM)	0.05~0.5µl	
Target RNA (10µM)	0.05~0.5µl	
ssRNA Reporter (FAM-BHQ1)	0.05~0.5µl	
Nuclease-free ddH <sub>2</sub> O	Up to 20µl	

Note:1)When the dosage is small, each component can be diluted first and then added to the system.

2)The crRNA, Target RNA, and ssRNA Reporter can be diluted with Nuclease free ddH<sub>2</sub>O, but for very low concentrations of Target RNA (such as LOD experiments), it is recommended to dilute with 0.1% Tween 20.

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

2. Real time fluorescence quantitative PCR instrument detects fluorescence signals, reacts at 37°C, and collects fluorescence

## For Research Use Only



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signals every 30 seconds.

## Note

- 1. Cis cleavage of Cas13a: Cas13a specifically cleaves target RNA under the guidance of crRNA, and does not have strict requirements for the PFS sequence of target RNA. Then use after high-temperature and high-pressure treatment. It is recommended to wear a disposable mask during operation.
- 2. The Cas13a protein and reaction buffer provided by our company do not contain any nuclease activity other than Cas13a.
- 3. Trans cleavage of Cas13a: When target RNA is present, Cas13a/crRNA forms a ternary complex with target RNA (Cas13a/crRNA/target RNA). At the same time, Cas13a is stimulated with trans cleavage activity, chopping up any sequence of single stranded RNA in the reaction system.
- 4. Cas13a protein is heat sensitive and prone to inactivation. A reaction system should be prepared on ice throughout the entire process, and the enzyme should be immediately stored at -20 °C after use.
- 5. To prevent RNase pollution, please keep the experimental area clean and tidy. Wear clean gloves and masks during operation, and use RNase free consumables such as gun heads and centrifuge tubes for the experiment.
- 6. This product is for scientific research purposes only and shall not be used for clinical medical diagnosis or other unreasonable purposes.