



LwaCas13a (C2c2) enhanced v2 Instructions

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Catalog Code: CAS-13-001
CAS-13-010
CAS-13-100

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Product Information

Product name	LwaCas13a enhanced v2*
Expression system	Escherichia coli
Quality	Recombinant protein
Form	Liquid
Molecular weight	138 kDa

Product Introduction

LwaCas13a protein is derived from *Leptotrichia wadei*. Cas13 forms a functional complex with crRNA, "gliding" over target RNA, and upon successful pairing, Cas13 is specifically activated, trans-cleaving the surrounding reporter molecule. This feature can be used in the field of molecular diagnostics to detect pathogens.

Storage

-20°C. Aliquot after receiving. Avoid repeated freeze-thaw.

Materials supplied

Cat:	CAS-13-001	CAS-13-010	CAS-13-100
LwaCas13a protein	10 μ M*20 μ l (200pmol)	10 μ M* 200 μ l (2,000pmol)	10 μ M *1000 μ l (10,000pmol)
10X Cas13a Reaction Buffer	1 ml * 1 vial	1 ml * 2vial	10 ml * 1vial
1X Diluent Buffer	1 ml * 1 vial	1 ml * 2 vial	5 ml * 1 vial

Other materials required

1. Reporter: substrate gives signals when digested by Cas13. Recommend EZassay™ biotech. products (Reporter (for Cas13) Cat.#. RNA-FAM-BHQ.)

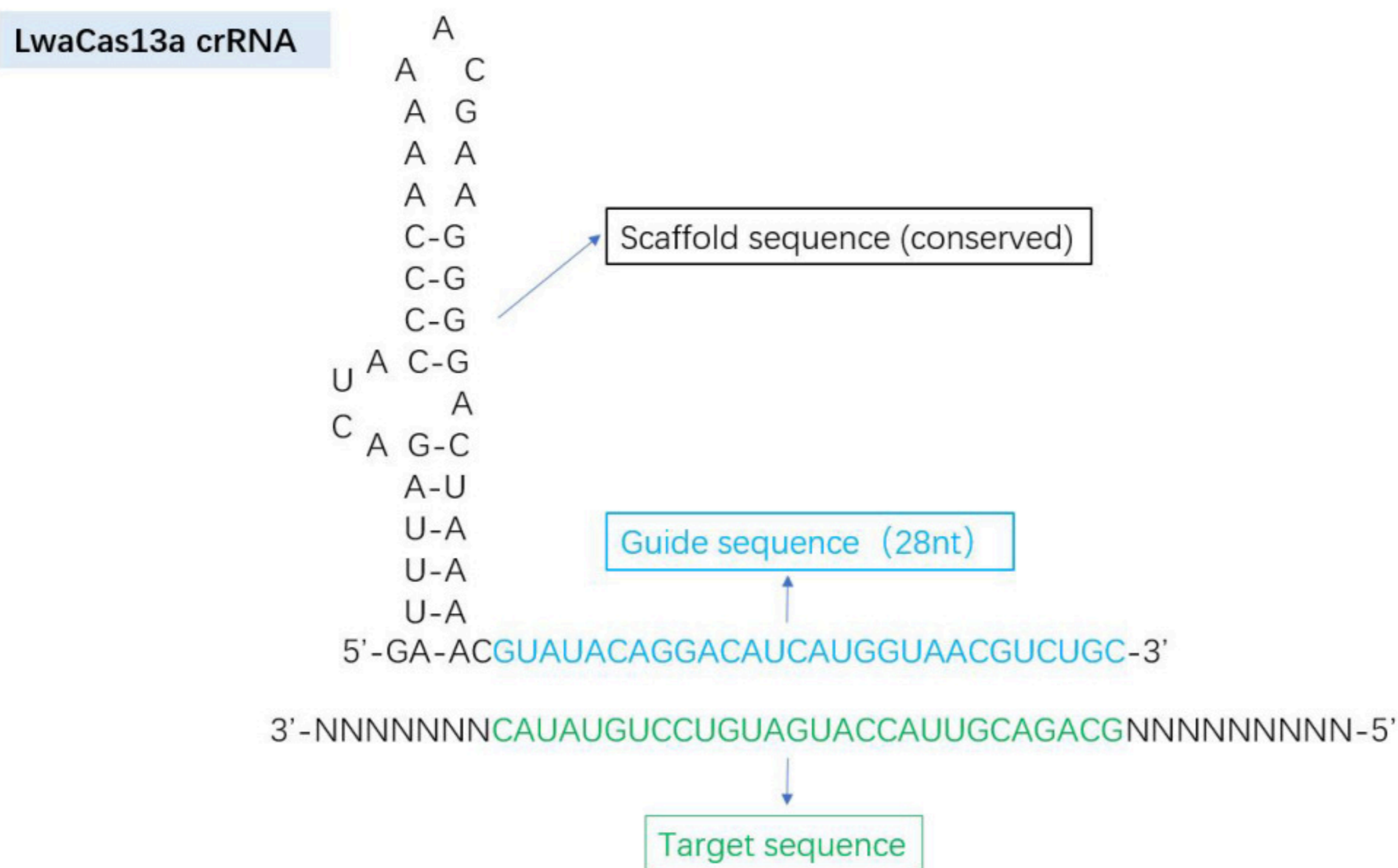
Cat:	Item
DNA-FAM-BHQ	Reporter DNA-FAM label
DNA-FAM-BIO	Reporter DNA-FAM-Biotin label

2. crRNA/gRNA:

LwaCas13a crRNA scaffold sequence:

5' - GAUUUAGACUACCCCAAAAACGAAGGGGACUAAAAC -3'

Any question about design, contact info@ezassay.com



Assay procedure

- 1、 Amplification of target DNA. (Optimal step)
- 2、 Prepare reaction as described in the table below.

Component	Recommend	Optimization
10×Cas13a Reaction Buffer	1×	-
10 mM NTPs	1 mM each	-
Reporter (FAM-BHQ1 labelled RNA)	120 nM	40 ~ 160 nM
crRNA	20 nM	20 ~ 80 nM
LwaCas13a enhanced v2*	100 nM*	40 ~ 160 nM
RNase Inhibitor	40 U total	-
T7 RNA Polymerase	30 U total	5 ~ 50 U
DNA amplicons (from 1st step) **	optimal	0-5ul
H2O	up to 20 µL	-

* LwaCas13a is provided as 1µM or 10µM. Please make dilution before use.

**Please makes sure there is T7 promoter in the amplification product.

- 3、 Set temperature at 37°C, read fluorescence every 1 min for 30~60 minutes.

Compare Cas13a enhanced v2 and Cas13a wild type



Positive control groups①&②: Cas13&crRNA complex was activated. Reporter was cleaved by Cas13 and fluorescence was detected. The activity of Cas13a_enhanced_v2 was 50% higher than Cas13a_wild type.

No template control group: Cas13&crRNA complex was inactivated. No fluorescent signal was detected.

Notes

1. This product is for scientific research only, please read this manual carefully before use.
2. Please strictly follow the management norms of gene amplification laboratory to conduct experimental operations and pay attention to prevent contamination of amplification products.
3. After the end of the experiment, the waste generated during the testing process shall be disposed of in accordance with relevant regulations.