EZassay

AsCas12a（Cpf1）Protein Instructions

info@ezassay.com

www.ezassay.com

EZassay Biotechnology Ltd.

Catalog Code: CAS-12A-000
CAS-12A-001
CAS-12A-010
CAS-12A-100

CONTENTS

Product Information	
Product name	AsCas12a protein
Expression system	Escherichia coli
Quality	Recombinant protein
Form	Liquid
Molecular weight	151 kDa

Product Introduction

AsCas12a protein is derived from Acidaminococcus sp. Cas12a forms a functional complex with crRNA, "gliding" over the amplification product of the DNA, and upon successful pairing, Cas12a 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-12A-000	CAS-12A-001	CAS-12A-010	CAS-12A-100
AsCas12a protein*	10μM*10μL (100pmol)	10μM*20μL (200pmol)	10μM* 200μL (2,000pmol)	10μM *1000μl (10,000pmol)
Cas12a Reaction buffer（10X）	1 ml * 1	1 ml * 1	1 ml * 4	10ml*2
Diluent Buffer (Cas12a) (1X)**	1 ml * 1	1 ml * 1	1 ml * 2	10 ml * 1

* AsCas12a protein is supplied in a buffer containing 50% glycerol.
** Diluent Buffer (Cas12a)(1X) is a protein dilution buffer containing 50% glycerol.
A glycerol-free version can be customized for lyophilization applications upon request.

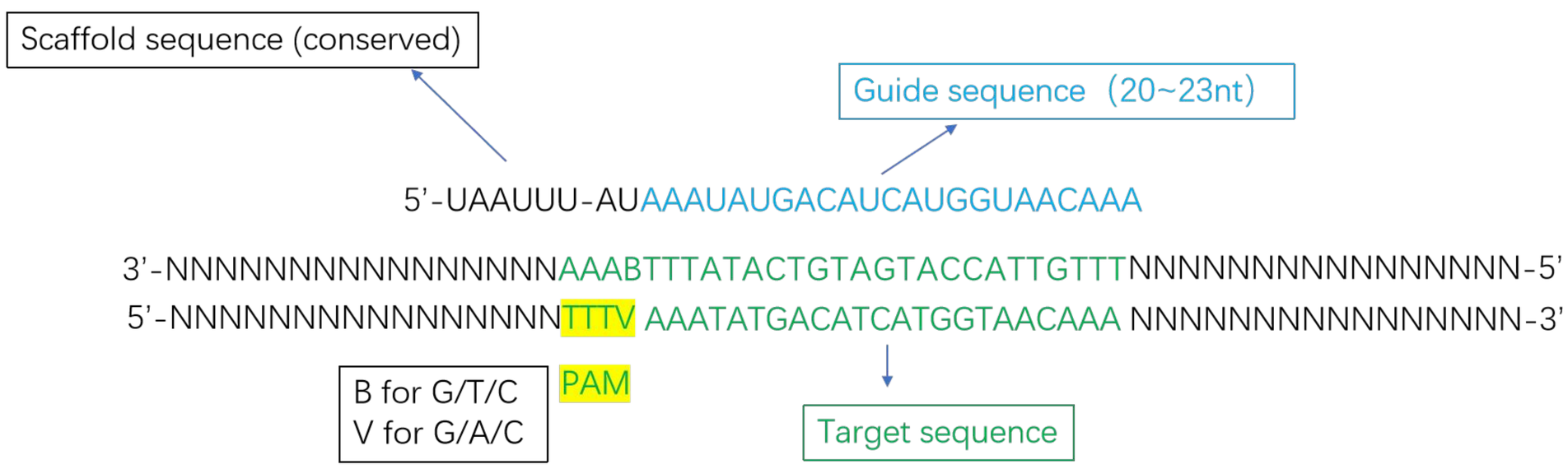
Other materials required

1. Reporter： substrate gives signals when digested by Cas12a. Self-design, for example FAM-BHQ labelled ssDNA. Optional select EZassay™ biotech. products (Reporter DNA FAM label (for Cas12a) Cat.#. DNA-FAM-BHQ.)

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

2. crRNA/gRNA：
AsCas12a crRNA scaffold sequence: 5’ -UAAUUUCUACUCUUGUAGAU-3’
If any question about design, please contact info@ezassay.com

AsCas12a crRNA



Assay procedure

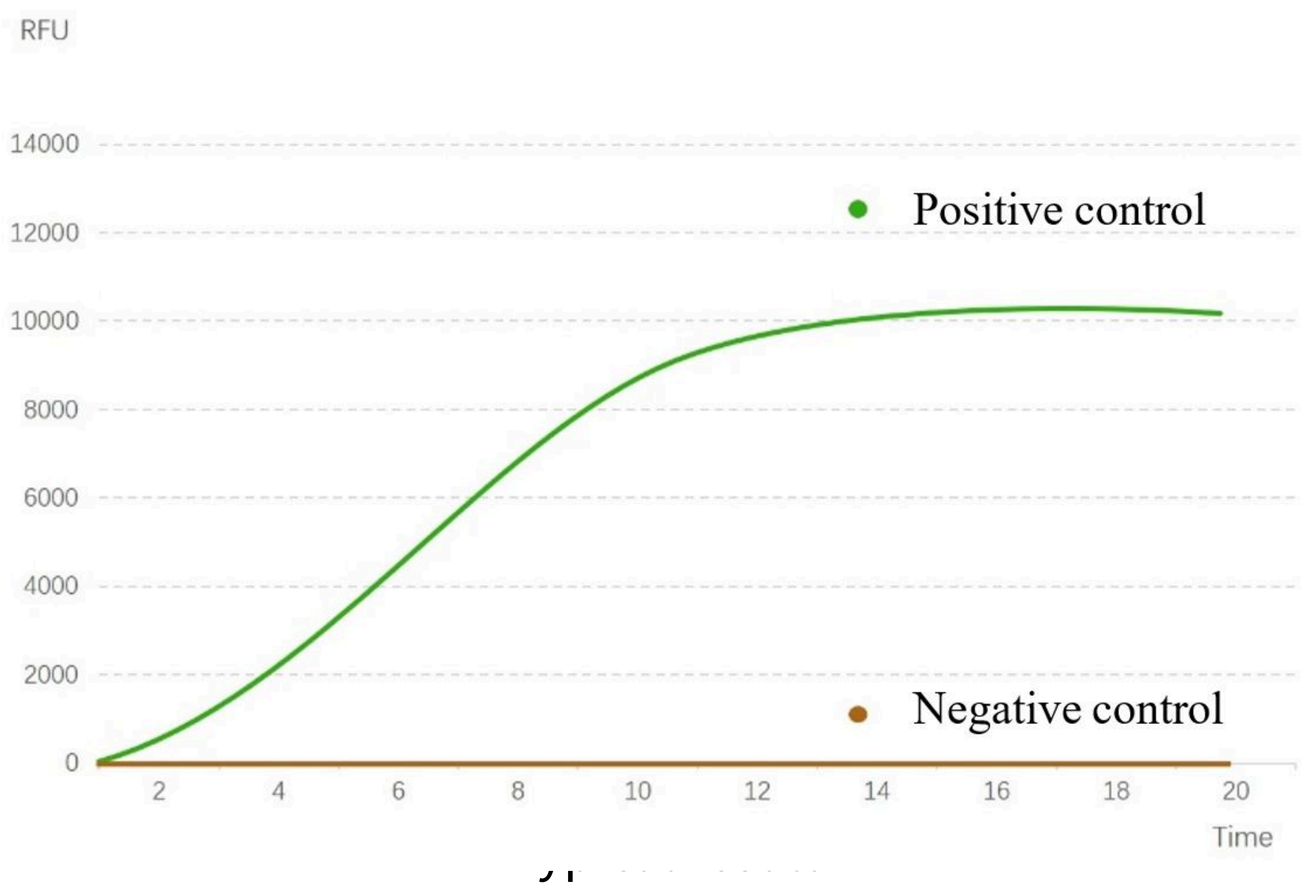
1、Amplification of target DNA. (reverse transcription firstly for RNA target) (Optimal step)

2、Prepare reaction as described in the table below.

Component	Recommend	Optimization
Cas12a Reaction Buffer(10X)	1×	-
Reporter (FAM-BHQ1 labelled ssDNA)	120 nM	40 ~ 200 nM
crRNA	50 nM	20 ~ 200 nM
AsCas12a protein*	20 nM	10 ~ 100 nM
DNA amplicons (from 1st step)	optimal	0-5ul
H2O	up to 20 µL	-

*The optimal enzyme usage varies among manufacturers due to differences in enzymatic activity and reaction buffer systems. Consequently, the ideal concentration of Cas12a must be determined on a per-project basis. For initial use, we recommend performing a comparative test across a concentration gradient ranging from 5 nM to 100 nM.

3、Set temperature at 37°C， read fluorescence every 1 min for 20~60 minutes.



Positive control: Cas12a&crRNA complex is activated by target DNA. Reporter, FAM-BHQ1 labelled ssDNA, is cleaved by Cas12a and fluorescence is released.
Negative control: Without target DNA, Cas12a&crRNA complex is inactivated. No fluorescent signal is 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.

Professional supplier of point-of-care test products

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