



BrCas12b Protein Instructions

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

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Product Information	
Product name	BrCas12b (from Brevibacillus leverages)
Expression system	Escherichia coli
Quality	Recombinant protein
Form	Liquid
Molecular weight	170 kDa

Product Introduction

BrCas12b is derived from Brevibacillus leverages. Cas12b and sgRNA form functional complex and it searches for target sequence. The trans-cleavage activity of Cas12b is activated when sgRNA binds target sequence. Cas12b efficiently cuts ssDNA around. Cas12b features resistance of high temperature of 65~70°C.

Storage

-20°C. Suggest aliquot after receiving. Avoid repeated freeze-thaw.

Materials supplied

Cat:	CAS-12R-000	CAS-12R-001	CAS-12R-010	CAS-12R-100
BrCas12b protein	10μM*10μL (100pmol)	10μM*20μL (200pmol)	10μM* 200μL (2,000pmol)	10μM *1000μl (10,000pmol)
Cas12b Reaction Buffer(10X)	1 ml * 1	1 ml * 1	1 ml * 4	10ml*2
Diluent Buffer (1X) (Cas12b)	1 ml * 1	1 ml * 1	1 ml * 2	10 ml * 1

Other materials required

1. Reporter: substrate gives signals when digested by Cas12b. Self-design, for example FAM-BHQ labelled ssDNA. Optional select EZassay™ products (Reporter DNA FAM label (for Cas12b) Cat.#. DNA-FAM-BHQ-B.)

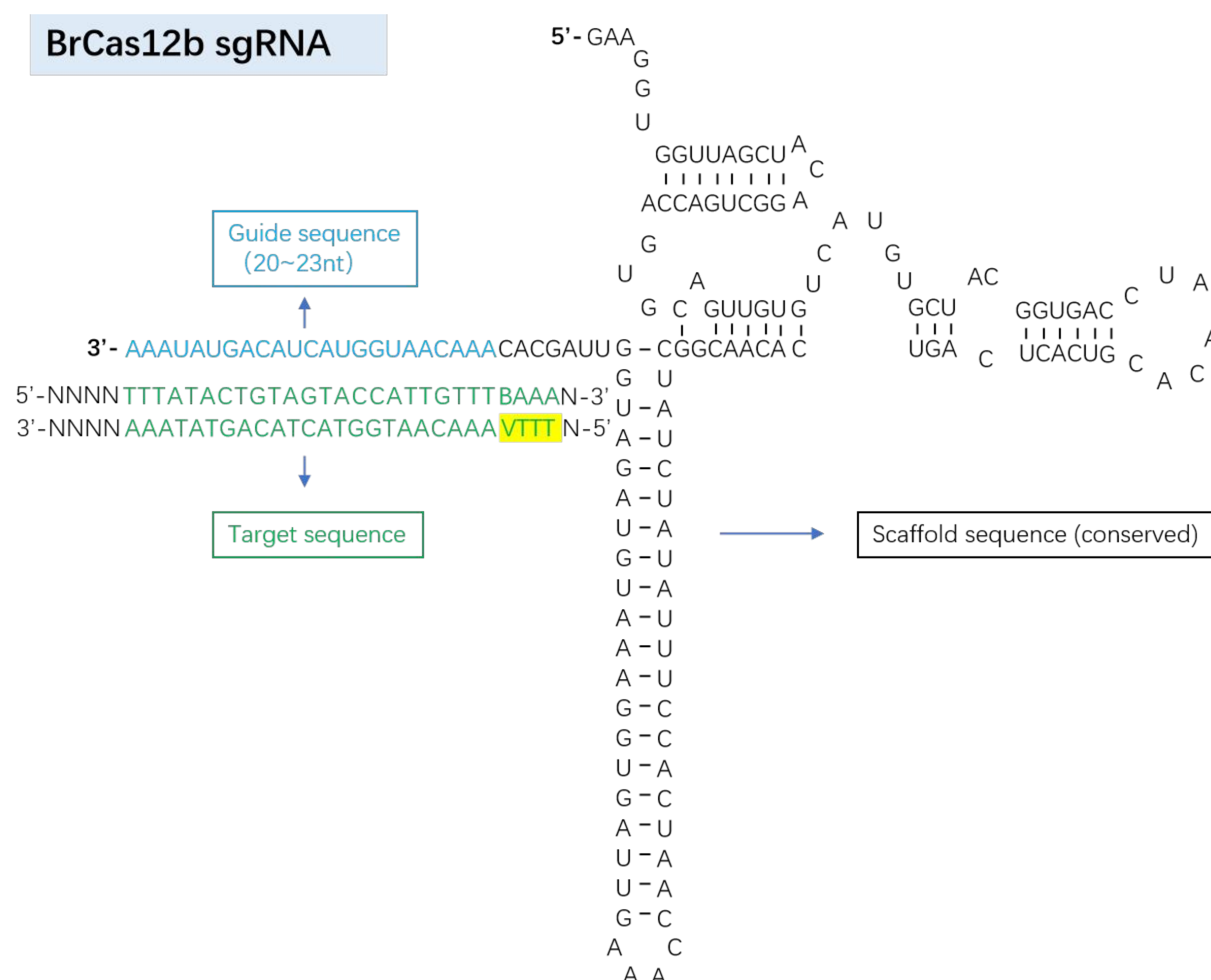
Cat:	Item
DNA-FAM-BHQ-B	Reporter DNA FAM label
DNA-FAM-BIO-B	Reporter DNA FAM-Biotin label

2. **sgRNA:** forms functional complex with BrCas12b. Recommend EZassay™ High Yield crRNA/sgRNA Synthesis Kit to prepare sgRNA. (Cat.#: SG-RNA-001)

(BrCas12b sgRNA scaffold sequence结构序列: 5' -

GAAGGUGGUUAGCUACAGGCUGACCAGUGCAGUUGUGUCAUGUGCUACGGUGACCUAACACG
UCACUCAGUCACAACGGCUAUCUAUAUUUCCACUAACCAAAGUUAGUGGAAAUGUAGAUGGU
UAGCAC -3')

Any question about design, contact info@ezassay.com.



3. DNA samples such as amplification products. Recommend EZassay™ LAMP isothermal amplification kit. (Cat.#.: LAM-LQ-1.)

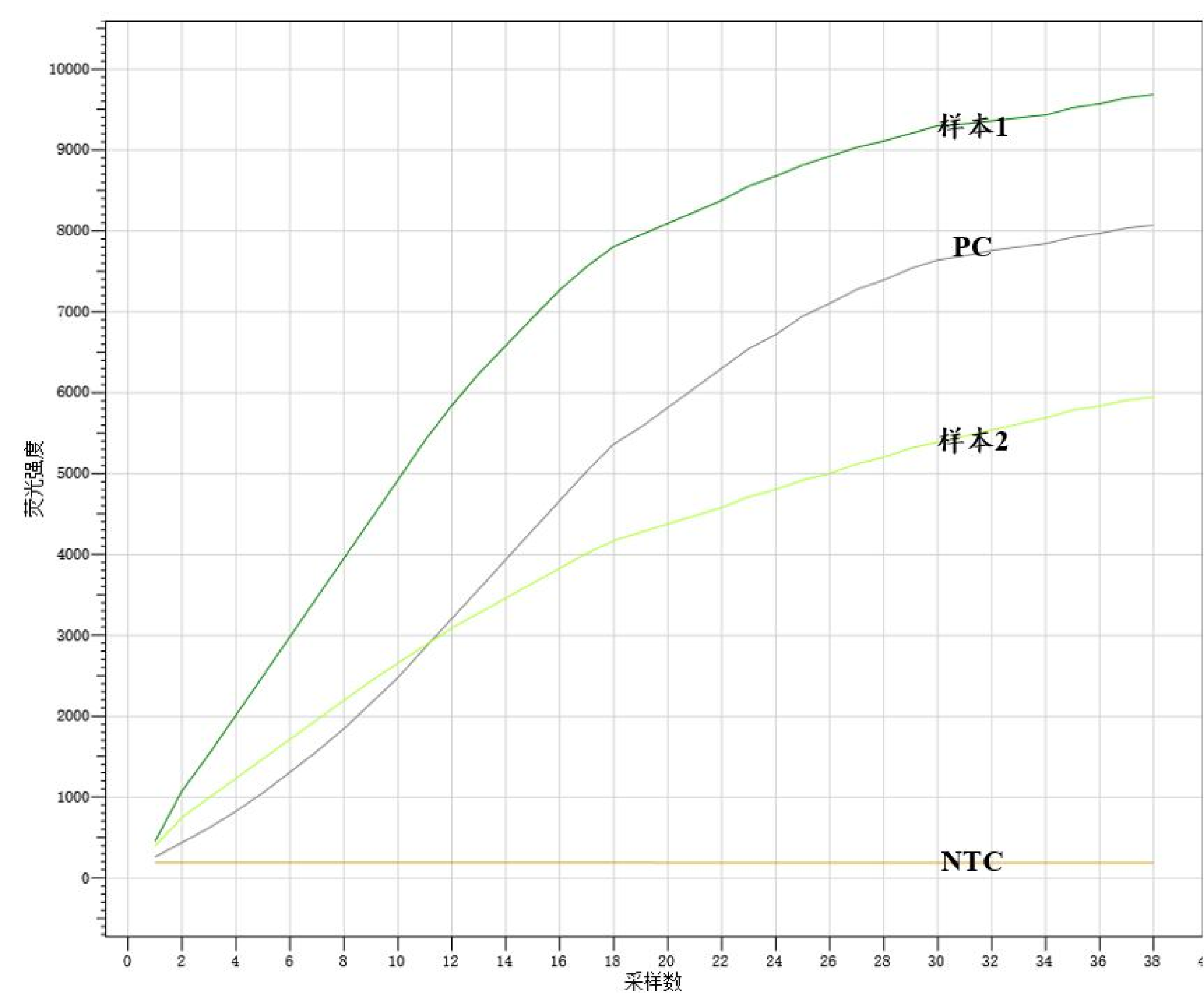
Assay procedure

- 1、Amplification of target DNA (reverse transcription firstly for RNA target) by LAMP.
(Optimal)
- 2、Prepare reaction as described in the table below.

Component	Recommend concentration	Optimization range
Cas12b Reaction Buffer (10X)	1×	-
Reporter ssDNA (fluorescence)	250 nM	50 ~ 500 nM
sgRNA	250 nM	200 ~ 1000 nM
BrCas12b	250 nM (=0.25uM)	200 ~ 500 nM (=0.2~0.5uM)
DNA sample (eg. amplification product)	Optimal	0-5ul
ddH2O	Up to 20 μL	-

Set temperature at 65°C, read fluorescence every 1 min for 30~40 minutes.

3、Typical result:



Positive control (PC): Test samples (sample1-2): Cas12b efficiently cuts reporter and gives strong fluorescence signal after activated by LAMP amplicons.

No template control (NTC): No fluorescence signal for no template LAMP amplification group.

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.