ACE Biolabs
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Datasheet

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Cas9 Nuclease

Cat# ER1006 – 50 pmole

Storage at -20 ℃

INTRODUCTION

Cas9 Nuclease is a RNA-guided, site-specific double strand DNA nuclease. Guided by a target-specific, single guide RNA (sgRNA), the Cas9 nuclease, with two cleavage active site, cleave both strands upon recognition of the target sequence by the sgRNA, resulting in double-stranded breaks. This product is a high purity Cas9 Nuclease from recombinant *Streptococcus pyogenes*.

CONTENTS

No	Component	**ER1006 – 50 pmole**
BA	Cas9 Nuclease	25 µl
BB	10X Cas9 Reaction Buffer	1 ml

UNIT DEFINITION

One unit (U) is defined as the amount of enzyme that required to make 0.5 pmole dNTP incorporate into acid-insoluble material in 30°C for 10 min.

REACTION CONDITIONS

1 × Cas9 Nuclease Reaction Buffer, incubate at 37°C.

QUALITY CONTROL

1. Protein Purity: The purity of Cas9 Nuclease is higher than 95%, detected by Coomassive blue staining.
2. RNase contamination test: Incubation of 40 ng RNA and 1 pmol Cas9 Nuclease in 10 µl Cas9 Nuclease reaction buffer at 37°C for 4 hr result in higher than 90% integrity of RNA after gel electrophoretic.
3. Exonuclease Activity: Incubation of 0.6 µg Supercoiled pBR322 DNA and 1 pmol Cas9 Nuclease at 37°C for 1 hour results in no detected change in DNA bands after gel electrophoretic.
4. Endonuclease Activity: Incubation of 1µg λHindIII and 1 pmole Cas9 Nuclease at 37°C for 4 hour results in no detected change in DNA bands after gel electrophoretic.

PROTOCOL

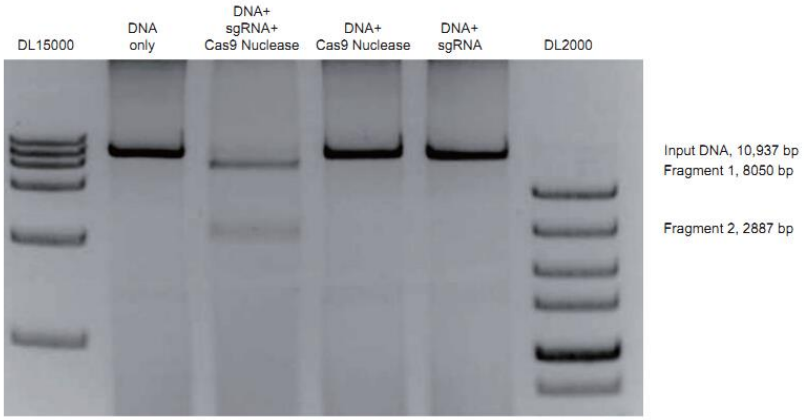
Note:

1. To ensure best cleavage quality, the molar ratio of Cas9 nuclease, sgRNA and target DNA should be 10:10:1 or higher. Dilute sgRNA and DNA with nuclease-free water into final concentration 300 nM and 30 nM.
2. To avoid RNase contamination, please wear mask and use nuclease-free reagent and tips

1. Prepare the reaction mixture in microcentrifuge tube as following:

10X Cas9 Reaction Buffer	3 µl
300 nM sgRNA	3 µl
1 µM Cas9 Nuclease	1 µl
Nuclease-free Water	To 27 µl

2. Incubate at 37°C for 10 min.
3. Add 3 µl DNA into mixture and mix gently.
4. Incubate at 37°C for 1 hour.
5. The products can be analyzed directly by 2% agarose gel electrophoresis.
6. Result:



PRODUCT USE LIMITATION

These products are intended for research use only.