Name	Cat#	Size	Concentration	Price
TtAGO nuclease	TtAGO-10	200 pmol, 200 µl	1 µM	$150.00
TtAGO nuclease	TtAGO-50	1 nmol, 200 µl	5 µM	$490.00

TtAGO (Tth Argonaute)

Description

Prokaryotic Argonaute (pAGO) is the pivotal enzyme in the host defense system by mediating nucleic acid molecules. As the DNA-guided endonuclease from eubacterium *Thermus thermophilus*, TtAGO protein targets cognate DNA without the restriction of a PAM (protospacer adjacent motif) or PFS (protospacer flanking site) on the target sequence, expanding the selection range of available target DNA sequence. Besides, compared with CRISPR/Cas systems, TtAGO have remarkable advantages as TtAGO endonuclease utilizes guide DNA rather than guide RNA in directing target DNA cleavage, which is more convenient for in vitro applications as guide DNA is more stable in ambient temperature, easier and cost-saving to synthesize than guide RNA.

These capacities enable TtAGO as a powerful programmable DNA endonuclease directed by short guide DNAs in efficient Nucleic Acid Detection systems as well as in versatile Synthetic Biology platforms.

Features

High Biological Activity

Highly Specific

Low Endotoxin Level

Application

Nucleic Acid Detection, Cancer Diagnostics, Genetic Screening, Synthetic Biology

Source

TtAgo gene from *Thermus thermophilus* (NCBI Reference Sequence: WP_011174533.1) expressed in E. coli

Purity

Greater than 95% as determined by SDS-PAGE and FPLC

Activity

Purified 370 bp PCR product dsDNA digested after 1 hour reaction at 83°C.

The reaction is performed as follow:

Reaction system total 10 µl, including 50 nM TtAGO Enzyme, 0.5 µM each of guide DNA, 100 ng purified 370 bp PCR product dsDNA as target DNA and 2ul 5x reaction buffer.

Reaction at 83°C for 1 hour, then incubate at 95°C for 10 min to terminate the reaction.

Storage Condition

-20 °C

5X Reaction Buffer

100 mM Tris-HCl, pH 8.8,

50 mM $(NH_4)_2SO_4$,

50 mM KCl,

40 mM $MgSO_4$,

0.5% Tween-20,

4 M Betaine

Storage Buffer

10 mM Tris-HCl

300 mM NaCl

1 mM DTT

0.1 mM EDTA

50% Glycerol

pH 7.4 at 25°C

Experimental Data

Fig.1

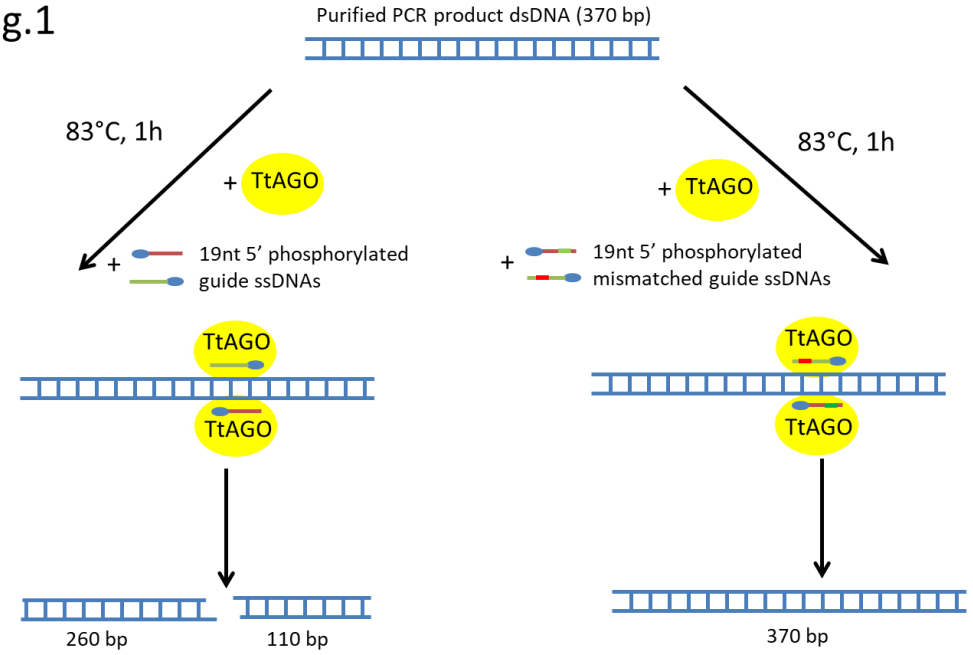

Figure 1. Schematic overview of the TtAGO-based ssDNA-directed DNA cleavage method.

Fig.2

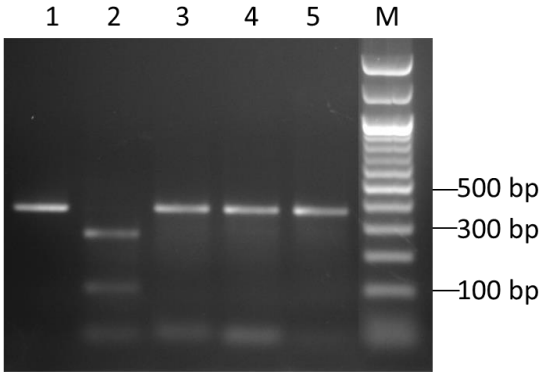

Figure 2. Agarose gel electrophoresis showing the TtAGO-based DNA cleavage results. DNA size showing as Figure 1.
Lane 1, 100 ng purified PCR product dsDNA.
Lane 2, 100 ng purified PCR product dsDNA + 50 nM TtAGO + 0.5 μM each of guide DNA.
Lane 3, 100 ng purified PCR product dsDNA + 50 nM TtAGO + 0.5 μM each of mismatched guide DNA; Mismatched site is on the position 11 of guide DNA.
Lane 4, 100 ng purified PCR product dsDNA + 50 nM TtAGO + 0.5 μM each of mismatched guide DNA; Mismatched site is on the position 12 of guide DNA.
Lane 5, 100 ng purified PCR product dsDNA +50 nM TtAGO + 0.5 μM each of mismatched guide DNA; Mismatched sites are on the positions 11 and 12 of guide DNA.
M, MCLAB 100 bp DNA ladder.

References

1. Gang Sheng et. al. (2014). PNAS. 111 (2) 652-657.

2. Daan C. Swarts et. al. (2014). Nature. 507(7491):258-261.

3. Yuqing Qin et. al. (2022). Trends Biotechnol. 40(8):910-914.