E.coli Poly(A) Polymerase

Cat.No.: HYJ233

Product Description

Poly(A) polymerase catalyzes the addition of AMP, converted from ATP, to the 3' end of RNA in a template-independent manner.

Components

Components	100 U	500 U
E. coli Poly(A) Polymerase (5,000 U/mL)	20 μL	100 μL
10X Poly(A) Polymerase Reaction Buffer	1.5 mL	1.5 mL
ATP(10 mM)	200 μL	200 μL

Storage

Store at -20°C.

Storage Buffer

20 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 1 mM EDTA, 50% Glycerol, 0.1% (w/v) Triton® X-100, pH 7.5 @ 25°C

Source

Derived from an Escherichia coli strain carrying the Poly(A) polymerase gene cloned from E. coli.

Activity Definition

One unit is defined as the amount of enzyme required to catalyze the incorporation of 1 nmol of AMP into RNA in 10 minutes at 37°C in a 20 μl reaction mixture.

Applications

- Labeling RNA with ATP or cordycepin.
- Adding Poly(A) tails to RNA for cloning or affinity purification.
- Increasing mRNA stability to enhance translation efficiency in eukaryotic cells after transfection.

Inhibition and Inactivation

Inactivate at 80°C for 15 min.

Instruction

1. Add the following reaction components on ice according to the table below (example for a 20 μL reaction volume):

Components	Volume
10X Poly(A) Polymerase Reaction Buffer	2μL
ATP(10 mM)	2μL
E. coli Poly(A) Polymerase	1μL
RNA（1-10 μg）	10μL
ddH_2O	Up to 20μL

2. Mix all reaction components thoroughly and briefly centrifuge to collect the solution at the bottom of the tube.
3. Incubate at 37°C for 30 minutes.
4. Heat inactivate by incubating at 85°C for 15 minutes or add EDTA to a final concentration of 10 mM.
5. RNAase inhibitor can be added to enhance RNA stability in the solution, with a 1X concentration of 1 U/μL.