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## Poly(A) Polymerase, Yeast

Store at -20°C

Cat. No.	Description	Concentration	Quantity
E017	Poly(A) Polymerase, Yeast	1 U/μl	100 U (100 μl)

### Product Description

Poly(A) Polymerase catalyses the template independent addition of adenosine residues onto the 3' ends of polyribonucleotides. The use of ATP as a substrate leads to poly(A) tailing whereas substitution of cordycepin-5'-triphosphate (3'-dATP) for ATP results in addition of a single dA residue to the 3'-termini of the RNA. Neither ADP nor dATP can be used as substrates for this enzyme. Poly(A) Polymerase from yeast has been shown to be more effective at oligonucleotide-labeling and poly(A) tailing of long RNA templates than Poly(A) Polymerase from *E. coli*.

### Product Components

Part No.	Product Components	100 U
E017-1	Poly(A) Polymerase, Yeast (1 U/μl)	100 μl
E017-2	5X Poly(A) Polymerase, Yeast Reaction Buffer	1 ml
E017-3	25 mM MnCl <sub>2</sub>	500 μl
E017-4	ATP (10 mM)	150 μl

### Product Applications

- Labelling of RNA with ATP or cordycepin
- Poly(A) tailing of RNA for cloning or affinity purification
- Increasing translation of RNA transferred into eukaryotic cells

### Product Source

Recombinant *E. coli*.

### Enzyme Storage Buffer

20 mM Tris-HCl (pH 8.0), 100 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 0.1% Triton® X-100 and 50% (v/v) Glycerol.

### Enzyme Unit Definition

One unit is defined as the amount of Poly(A) Polymerase, Yeast that catalyzes the incorporation of 1 nmol of AMP into RNA in 10 minutes at 37°C.

### Storage Conditions

Store all components at -20°C. Avoid repeated freeze-thaw cycles of all components to retain maximum performance. All components are stable for 1 year from the date of shipping when stored and handled properly.

### 5X Poly(A) Polymerase, Yeast Reaction Buffer Components

250 mM Tris-HCl, 1 M NaCl, 50 mM MgCl<sub>2</sub>, 5 mM DTT, pH 8.

### Protocol

#### 3'-End labelling of RNA

1. Add the following components to a sterile tube sitting on ice:

Components	Volume	Final Concentration
RNA	Variable	0.2 μM
Cordycepin-5'-Triphosphate	Variable	0.4 μM
Poly(A) Polymerase, Yeast (1 U/μl)	1 μl	1 U
5X Poly(A) Polymerase, Yeast Reaction Buffer	2 μl	1X
25 mM MnCl <sub>2</sub>	1 μl	2.5 mM
Nuclease-free H <sub>2</sub> O	up to 10 μl	-

2. Collect all components by a brief centrifugation. Incubate the reaction at 37°C for 10 minutes.

3. The 3'-End labelled RNA product is ready for immediate downstream applications or for long-term storage at -80°C.

#### Poly(A) tailing of RNA

1. Add the following components to a sterile tube sitting on ice:

Components	Volume	Final Concentration
RNA	Variable	0.2 μM
ATP (10 mM)*	1.25 μl	0.5 mM
Poly(A) Polymerase, Yeast (1 U/μl)	1 μl	1 U
5X Poly(A) Polymerase, Yeast Reaction Buffer	5 μl	1X
25 mM MnCl <sub>2</sub>	2.5 μl	2.5 mM
Nuclease-free H <sub>2</sub> O	up to 25 μl	-

\*Radiolabelled, biotinylated or fluorescently-labelled ATP can be substituted in the reaction.

2. Collect all components by a brief centrifugation. Incubate the reaction at 37°C for 10 to 20 minutes.

3. Terminate the reaction by heating at 65°C for 20 minutes or by adding 5 mM EDTA.

4. The Poly(A)-tailed RNA product is ready for immediate downstream applications or for long-term storage at -80°C.

#### Heat Inactivation

65°C for 20 minutes.