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AMV Reverse Transcriptase

Product Number: RT06

Shipping and Storage

Store at -80°C. If stored for a short period of time, it can be stored at -20°C. (Repeated freeze-thaw activity may decrease, please try to avoid it as much as possible). Transportation conditions: Dry ice transportation.

Components

Component	RT06	RT06
AMV Reverse Transcriptase(10U/µl)	200U	1KU

Description

AMV Reverse Transcriptase was isolated from avian myeloblastoma virus (AMV) using a method improved by Houts et al. The isolated enzyme has a molecular weight of 157KD $\alpha\beta$ Whole enzyme. This enzyme is highly purified and completely free from nuclease contamination. This enzyme can perform cDNA synthesis using approximately 10 kb RNA template. This enzyme has both cDNA synthesis activity and RNase H activity.

Unit definition

Using Poly (rA)·Oligo (dT) as a template/primer, the amount of enzyme required to introduce 1 nmol of [3H] dTTP into acid insoluble precipitate within 10 minutes at 37 °C is defined as 1 active unit (U).

Note

The suitable reaction temperature for this enzyme is 42-58°C. When the temperature of cDNA synthesis reaction exceeds 50°C, it is recommended to first take a 4-minute warm bath at 42°C before proceeding with the reaction above 50°C.

Protocol

1. Thaw the components on ice, mix the following components, and incubate at 42°C for 1 hour. If using a random primer mixture, it is recommended to incubate at 25°C for 5 minutes before incubation at 42°C.

Component	Volume(µL)	
RNA template	1ng-1µg total RNA or 50 pg-100ng poly(A)-RNA	
d(T) ₂₃ VN (50µM) or Random Primer Mix (60µM)	2	
10X AMV buffer	2	
AMV (10U/µl)	0.2 to 2	
10 mM dNTP Mix	1	
RNase Inhibitor (40U/µl)	0.2	
Nuclease-free H ₂ O	Up to 20	

Note: The usage range of AMV Reverse Transcriptase is 42-50°C.

2. Deactivate the enzyme at 85°C for 5 minutes. For downstream PCR applications, the volume of cDNA products should not exceed 1/10 of the PCR reaction volume.