Warm start M-MLV Reverse Transcriptase M2

Cat.No.: HYA214

Product Description

Warm start Reverse Transcriptase M2 is a highly active reverse transcriptase obtained through the rational screening platform-directed transformation based on Warm start Reverse Transcriptase. The enzyme has removed RNase H activity and has higher temperature tolerance. This product has been modified by hot start to block the reverse transcriptase activity at room temperature, which can effectively eliminate the adverse effects of RNA advanced structure and non-specific factors on cDNA synthesis, and has higher stability and reverse transcription synthesis ability.

Components

1. 200 U/μL Warm start Reverse Transcriptase M2
2. 5×WS Reverse Transcriptase buffer(optional)

*5×WS Reverse Transcriptase buffer does not contain dNTPs. Please add dNTPs when preparing the reaction system.

Storage

Store at -25°C~ -15°C.

Unit Definition

1 activity unit (U) is defined as the amount of enzyme required to convert 1 nmol of dTTP into acid-insoluble substances at 37°C for 10 min using Poly (A)·Oligo $(dT)_{25}$ as template/primer.

Instructions

1. Refer to the table below to prepare the system

Components	Volume
Template RNA	total RNA ≤ 5 μg；mRNA ≤ 1 μg
Oligo$(dT)_{12\text{-}18}$ Primer*	50 pmol
Or Random Primer *	50 pmol（20~100 pmol）
Or Gene specific Primer *	2 pmol
dNTPs（10 mM each）	1 μL

Nuclease-free Water	Up to 10 μL

*Choose different types of primers according to experimental needs.

2. Heat at 65℃ for 5 min, and quickly cool on ice for 2 min;
3. Add the following components to the above system to a total volume of 20 μL, and mix gently.

Components	Volume
5×WS Reverse Transcriptase buffer	4 uL
200 U/μL Warm start Reverse Transcriptase M2	1 uL
40 U/μL RNase Inhibitor	1 uL
Nuclease-free Water	Up to 20 μL

4. Perform the reaction under the following conditions.

1) If using Random Primer, incubate at 25℃ for 10 min first; then incubate at 50℃ for 30~60 min;
2) If using Oligo dT or specific primers, incubate at 50℃ for 30~60 min.

5. Inactivate Warm start Reverse Transcriptase M2 by heating at 95℃ for 5 min and terminate the reaction.
6. The reverse transcription product can be used directly for PCR reaction and real-time PCR reaction, or stored at -20℃ for a long time.

Technical Description

1. Suitable for reverse transcription temperature optimization in the range of 42℃~55℃.
2. With better stability, it is suitable for high-temperature reverse transcription amplification, which is conducive to effectively passing through the complex structure area of RNA and suitable for one-step multiplex fluorescence quantitative RT-PCR detection.
3. It has good compatibility with various PCR amplification enzymes and is suitable for high-sensitivity RT-PCR reactions.
4. It is suitable for cDNA library construction.
5. 3’ and 5’ RACE.