

M-MuLV Reverse Transcriptase

Cat. No: FPLF011.0025 FPLF011.0050

Contents:

| Components | 25 RXN | 50 RXN |
|---|--------|--------|
| M-MuLV Reverse Transcriptase Enzyme aliquoted from Thermo Fisher Co. | 25 μl | 50 μl |
| 5X RT Buffer | 100 µl | 200 µl |

Description:

This is a genetically modified RNA-dependent DNA polymerase requiring a DNA primer and an RNA template to synthesize a complementary DNA strand. Thermo-resistant H Minus M-MuLV Reverse Transcriptase has no RNase H activity. Therefore, degradation of RNA does not occur during firststrand cDNA synthesis, resulting in higher yields of full-length cDNA from long templates compared to other reverse transcriptases. Thermo-resistant H Minus M -MuLV Reverse Transcriptase maintains activity over a wide temperature range (42-52°C) which makes it an ideal tool for reverse transcription of RNAs having a high degree of secondary structure.

Kit storage:

This kit should be stored at -20 °C. Under this condition, reagents are stable for one year from the date of production.

 Mix the template RNA (total RNA or Poly (A) mRNA) and the primer in an RNasefree tube as below table. Optimal reaction conditions, such as the amount of RNA and primers, may vary and must be individually determined. Random hexamer or oligo (dT) 16 or specific primers could be used as primers.

| The concentration of template RNA and prime |
|---|
|---|

| Template RNA | Total RNA or Poly(A)+ mRNA | 10 ng~5 μg 5 ng~0.5 μg |
|--------------------|--------------------------------------|---------------------------|
| Primer | Oligo (dT)16 or Random hexamer | 1-2 μL 1 μL |
| DEPC-treated water | | Up to 13 µL* |

2) Incubate the mixture at 65 °C for 5 min chill on crash ice and add the reagent as follows:

* If you are using an RNase inhibitor, bring the upper solution to 12 μL and add 1 μL RNase inhibitor 20 U/ul

| Component | Volume (µL) |
|---------------------|-------------|
| 5X RT Buffer | 4 |
| 10 mM dNTP Mix | 2 |
| Thermo-Resistant RT | 1 |

- Mix by pipetting gently up and down (total reaction volume 20 μL).
- 4) Incubate for 10 min at 25 °C (omit this for Oligo dt).
- 5) Incubate for 60 min at 47 °C.
- 6) Stop the reaction by heating at 70 °C for 10 minutes. Chill on ice.

Disclaimers and Addresses:

This product is for **Research use only** and should only be used by trained professionals.