

Ultra-TM Thermo-resistant H Minus M-MuLV Reverse Transcriptase

(50 μ L, 200 U/ μ L, Total 10000 U) Store at -20 °C

Component	Volume
Ultra-TM Thermo-resistant RT	50 μΙ
5X RT Buffer	500 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. Ultra-TM Thermo-resistant H Minus M-MuLV Reverse Transcriptase has no RNase H activity. Therefore, degradation of RNA does not occur during first strand cDNA synthesis, resulting in higher yields of full-length cDNA from long templates compared to other reverse transcriptases. Ultra-TM Thermoresistant H Minus M-MuLV Reverse Transcriptase maintains activity over a wide temperature range (50-60 °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.

Protocol (first strand cDNA synthesis):

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

* If you use RNase inhibitor

Concentration of template RNA and primer			
Template	Total RNA	10 ng~1 μg	
RNA	or		
	Poly(A)+ mRNA	5 ng~0.5 μg	
Primer	Oligo (dT)16	1-2 μL	
	or		
	Random hexamer	1 μL	
DEPC-treated water Up to 12 uL (11 uL*)		to 12 uL (11 uL*)	

- 2- Incubate the mixture at 65 °C for 5 min and chill on crash ice and add the reagent as follow:
- 3- Mix by pipetting gently up and down (total reaction volume 20 μ L).
- 4- Incubate 10 min at 25 °C (omit this for Oligo dt).
- 5. Incubate 30 min at 55 °C.
- 6. Stop the reaction by heating at 80 $^{\circ}$ C for 5 minutes. Chill on ice.

Concentration of template RNA and primer		
5X RT Buffer	4	
RNase Inhibitor 20 U/μL (optional)	1	
10 mM dNTP Mix	2	
Ultra-TM Thermo-Resistant RT	1	

Disclaimers and Addresses

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

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