vivantis

DNA AMPLIFICATION PRODUCT

Product Datasheet

Product No : RTMM01
Quantity : 100 reactions

2X OneStep *Taq* ReverseTrans PCR Master Mix

Taq RTMM

Lot :
Expiry Date :
Supplied with : 1ml **2X OneStep *Taq* ReverseTrans PCR Master Mix** *
2ml of Nuclease-free Water

Store at -20°C

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Description :

2X OneStep *Taq* ReverseTrans PCR Master Mix offers rapid and sensitive end-point detection of RNA templates in a single step. 2X OneStep *Taq* ReverseTrans PCR Master Mix is an optimized ready-to-use 2X concentrated RNA amplification mixture containing M-MuLV Reverse Transcriptase, RNase Inhibitors, *Taq* DNA Polymerase, reaction buffer and dNTPs. It contains all the components required for routine RNA amplification except template and primers. Moloney Murine Leukemia Virus (M-MuLV) Reverse Transcriptase has the absence of RNase H activities that enhance the synthesis of long cDNAs and amplification of long transcripts. 2X OneStep *Taq* ReverseTrans PCR Master Mix allows one-step RT-PCR using only gene-specific primers.

Features :

- Saves time and reduces contamination due to reduced number of tests and pipetting steps.
- Stable at 4°C for 6 months, allowing immediate reaction setup without the time-consuming thawing of reagent.
- Suitable for all routine RNA amplification applications.

Storage and Stability :

- 2X OneStep *Taq* ReverseTrans PCR Master Mix is stable at -20°C for one year or 4°C for 6 months if properly stored.
- 2X OneStep *Taq* ReverseTrans PCR Master Mix is stable for 20 freeze-thaw cycles. To avoid frequent freeze-thaw, keeping small aliquot at -20°C is recommended.
- For daily use, keeping an aliquot at 4°C is recommended.

Quality Control :

All preparations are assayed for contaminating endonuclease, exonuclease, and non-specific RNase activities. Functionally tested in DNA amplification.

Recommended Protocol for 2X OneStep *Taq* ReverseTrans PCR Master Mix:

Gently mix all solutions after thawing. Spin down briefly and keep on ice.
Add the following components in a 0.2ml thin walled PCR tube on ice.

For 20µl reaction volume:

Reagent:	Volume	Final Concentration
Primers (Fwd / Rev)	Variable	0.1 - 1µM each
RNA template	Variable	0.02 - 5µg
Water, nuclease free	Adjust final volume to 10µl	
2X OneStep *Taq* ReverseTrans PCR Master Mix	10µl	*1X

* 40U M-MuLV Reverse Transcriptase, RNase Inhibitors, 1.5U Taq DNA Polymerase, 1X PCR Buffer, 0.2mM dNTPs Mix, and enhancers.
**Higher reaction volume may be achieved provided that the same final concentration of each reaction component is maintained. The number of tests will be reduced if the reaction volume more than 20µl.

Incubate primer-RNA template mix at 65 °C for 5 min before adding 2X OneStep *Taq* ReverseTrans PCR Master Mix.

Cycling Conditions (100bp - 500bp)	
cDNA Synthesis	42°C for 10 - 30 minutes
Initial Denaturation	94°C for 2 minutes
Denaturation	94°C for 15 seconds
Annealing	50 -68°C for 1 minute
Final Extension	72°C for 5 minutes

25 -40 cycles

*This protocol may change depending on the template RNA and primers used.

Cycling Conditions (300bp - 5kb)	
cDNA Synthesis	42°C for 10 - 30 minutes
Initial Denaturation	94°C for 2 - 5 minutes
Denaturation	94°C for 30 seconds
Annealing	50 -68°C for 30 seconds to 1 minute
Extension	72°C for 30 seconds to 2 minutes
Final Extension	72°C for 5 minutes

25 -40 cycles

Product Use Limitation
This product is for research purpose and *in vitro* use only

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