All-in-One First-Strand Synthesis MasterMix

REF: EG20131S

Storage Condition

-20℃

Components

Component	Amount
All-in-One First-Strand Synthesis MasterMix	400 μl
Nuclease-Free Water	2×1 ml

Description

All-in-One First-Strand Synthesis MasterMix is a high-quality, efficient, and convenient one-step cDNA synthesis mix. It is designed to minimize contamination and contains all the necessary components for first-strand cDNA synthesis, including thermostable M-MLV GIII Reverse Transcriptase and its reaction buffer, RNase inhibitor, dNTPs, Oligo(dT)$_{20}$VN, and random primers-all the necessary components. Simply add RNA template and water to initiate the reaction. Using the reverse transcription pre-mix, cDNA of up to 12 kb in size can be obtained within 15 minutes.

Protocol

1. Prepare the following reaction mixture on ice:

Reagent	Amount
Template RNA[a]	50 ng~1 μg
All-in-One First-Strand Synthesis MasterMix	4 μl
Nuclease-Free Water	To 20 μl

a. It is recommended to use high-quality RNA extracted using a kit, which has been treated to remove genomic DNA contamination, as the template for the reaction.

② Mix gently and spin down.

③ Incubate at 55℃ for 15 minutes.

Note: If the template RNA does not contain a poly(A) tail, you may pre-incubate at 25℃ for 10 minutes.

④ Terminate the reaction by incubating at 85℃ for 5 minutes.

⑤ Place the obtained cDNA product on ice for subsequent experiments.

Notice

The premix contains Oligo(dT)$_{20}$VN and random primers, suitable not only for eukaryotic mRNA containing poly(A) tail, but also for templates that do not contain poly(A) tail, such as prokaryotic RNA, eukaryotic rRNA, tRNA, etc. However, it is not suitable for small RNA like miRNA.

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