



miRNA All-In-One cDNA Synthesis Kit

Cat. No. G898

Store at -20°C.

Product Description

The **miRNA All-In-One cDNA Synthesis Kit** provides a comprehensive system for the efficient synthesis of first-strand miRNA cDNA from an RNA sample. The kit includes all the necessary components for a hassle-free setup, enabling RNA molecules to undergo polyadenylation and reverse transcription in a simple, single-tube reaction. This process utilizes Poly(A) Polymerase, OneScript® Plus Reverse Transcriptase, and a modified Oligo d(T) with Adapter that anneals to the poly(A)-tailed miRNA. To ensure RNA template integrity, the included SuperMix contains a recombinant RNaseOFF Ribonuclease Inhibitor, which protects RNA from degradation.

The synthesized first-strand miRNA cDNA can be directly used as a template for qPCR-based miRNA expression analysis with BlasTaq™ qPCR MasterMix (available separately). Users can select a suitable primer set from **abm**'s extensive collection of miRNA qPCR primers, which cover all known miRNA sequences. Each primer set contains a unique miRNA Forward Primer and a Universal 3' miRNA Reverse Primer that binds to the complementary Adapter sequence on the modified Oligo d(T).

Product Component	Quantity	Part No.
2X miRNA cDNA Synthesis SuperMix	20 rxn (200 µl)	P898-1
Enzyme Mix	20 rxn (40 µl)	P898-2
Nuclease-Free H ₂ O	1.0 ml	P100

Protocol

Reactions should be assembled in an RNase-free environment. The use of "clean" pipettors designated for PCR and aerosol-resistant barrier tips are recommended.

1. Thoroughly thaw and mix individual components before use and assemble reaction on ice.

Product Component	Volume
2X miRNA cDNA Synthesis SuperMix	10 µl
RNA Sample	Variable (200 ng small RNA or 2 µg total RNA)
Enzyme Mix	2 µl
Nuclease-Free H ₂ O	up to 20 µl

2. Gently mix the reaction and briefly centrifuge.
3. Incubate the mixture at 37°C for 30 minutes, followed by 50°C for 15 minutes. Immediately stop the reaction by heating at 85°C for 5 minutes. Chill on ice.
4. Newly synthesized first-strand miRNA cDNA can be directly used as a template for immediate downstream applications, or stored long-term at -20°C.

General Notes

- Both small RNA and total RNA can be used for first-strand cDNA synthesis; small RNA may give higher yields and improved purity of final products.
- RNA samples must be free of genomic DNA contamination.
- The first strand of miRNA cDNA can directly be used as a template for the qPCR-based analysis of miRNA expression. BlasTaq™ qPCR MasterMix (**Cat. No. G891**) is designed to work with our miRNA primer sets, maintaining optimal qPCR reaction conditions.
- The user can choose a suitable miRNA qPCR forward primer from **abm**'s comprehensive library. Included with this chosen forward primer is the Universal 3' miRNA Reverse Primer which binds to the complementary adapter sequence on the Oligo d(T).
- The Universal 3' miRNA Reverse Primer is also available separately as **Cat. No. MPH00000**.
- A variety of housekeeping gene primers for reaction controls are available including: U6 (**Cat. No. MPH00001**), SNORD44 (**Cat. No. MPH00003**), and SNORD48 (**Cat. No. MPH00005**).