



## SuScript cDNA Synthesis Kit

| Catalog No | Size              |
|------------|-------------------|
| • RT01A025 | 25 Rxn (250 µl)   |
| • RT01A026 | 100 Rxn (1000 µl) |

### Description:

SuScript cDNA Synthesis Kit is a cDNA synthesis kit containing all the components necessary to obtain cDNA in a high yield from RNA samples. The RNase Inhibitor presented in the kit adequately protects RNA templates against degradation. Optimized reaction buffer, MgCl<sub>2</sub>, Oligo-dTs, random hexamers and reverse transcriptase are also included in the 2X RTMix. The first strand of cDNA can be directly used as a template in downstream applications (PCR, Real-Time PCR, etc).

### Kit Components:

| Components             | RT01A025 | RT01A026 |
|------------------------|----------|----------|
| 2X RT Mix              | 2500 µl  | 1000 µl  |
| DNase/RNase Free Water | 250 µl   | 1000 µl  |

### General Considerations:

- High purity template RNA is essential for reliable efficient cDNA synthesis. A A260 / A280 ratio of 1.7 or higher is strongly recommended.
- The amount of template RNA is depended on the expected copy number of the sequence of interest. In general, 1 µg – 1 ng of total RNA is recommended, 0.05 – 100 ng if you are working with isolated mRNA.
- This protocol recommends cDNA synthesis for 1 hour at 37°C.
- To enhance the template coverage, the 5X RTMix also contains random hexamer primers. This provides multiple priming sites along the RNA for the detection of multiple short sequences.

### Recommended Protocol:

This protocol is exemplary for one reaction, and for multiple reactions it is necessary to calculate the components in a proportional manner. Mix the kit components in a micro tube in the recommended proportions below. Briefly centrifuge the tube to spin down the contents and to eliminate any air bubbles. Place the tube on ice until to reverse transcriptase reaction.

| Reaction Components    | Volume      |
|------------------------|-------------|
| 2X RT Mix              | 10 µl       |
| Total RNA              | 1-6 µl      |
| DNase/RNase Free Water | Up to 20 µl |
| Total Volume           | 20 µl       |

Mix gently and carry-out reverse transcriptase reaction conditions:

| RT Steps        | Temp (°C) | Time   | Cycle |
|-----------------|-----------|--------|-------|
| RT Reaction     | 37        | 1 hour | 1     |
| RT Deactivation | 80        | 10 min | 1     |

### For miRNA StemLoop miRNA RT Please follow:

| Reaction Components            | Volume      |
|--------------------------------|-------------|
| 2X RT Mix                      | 10 µl       |
| Total RNA                      | 1-6 µl      |
| Stem Loop (ST) Primer (10 µM ) | 1 µl        |
| DNase/RNase Free Water         | Up to 25 µl |
| Total Volume                   | 25 µl       |

Mix gently and carry-out reverse transcriptase reaction conditions:

| RT Steps        | Temp (°C) | Time   | Cycle |
|-----------------|-----------|--------|-------|
| RT Reaction     | 37        | 1 hour | 1     |
| RT Deactivation | 70        | 10 min | 1     |

### Notice:

The synthesized cDNA can be used immediately, without purification, or stored at -20°C for future use.

### Storage Conditions:

Store all contents at -20 °C in a freezer.

### Quality Control:

Nicking activity, priming activity, exonuclease activity, or endonuclease activity has not been detected.

