

# 4× EZscript Reverse Transcription Mix II

Catalog No.: EZB-RT2

## Description

The **EZBioscience® 4× EZscript Reverse Transcription Mix II** is a new-generation reverse transcription kit with higher reverse transcription efficiency compared to previous generations.

The kit is a ready-to-use premixed reagent that contains all the components necessary to synthesize first strand cDNA from total or poly(A)+ RNA using reverse transcriptase. Reaction products are applicable to subsequent PCR, qPCR. The mix contains reverse transcriptase, RNase Inhibitor, optimized buffer system, dNTPs; Oligo dT18 and Random Hexamer as primer. The random primer in the Mix can help get better Ct values.

The reverse transcriptase in this Mix is a genetic engineered enzyme based on M-MLV (RNase H-) reverse transcriptase. The reverse transcriptase lacking RNase H activity is suitable for preparing full-length cDNA. And the multiple site-mutations of reverse transcriptase can obviously increase its affinity to RNA templates and its strand extending ability, which make reverse transcription reaction more efficient. Moreover, this reverse transcriptase is rather resistant to common reverse transcriptase inhibitors. At the same time, the kit uses the latest optimized reaction system to further improve the reverse transcription efficiency. This product is also very suitable for reverse transcription using plant RNA.

When using this kit, 4× EZscript RT Mix II should be added to total RNA template, as recommended in the protocol, and add ddH<sub>2</sub>O to a total volume of 20 µl. Then, mix well and run the RT reaction.

## Components

Components	EZB-RT2 (100 Rxns)	EZB-RT2-L (500 Rxns)
4× EZscript RT Mix II	550 µl	550 µl × 5 tubes
Nuclease free ddH <sub>2</sub> O	1 ml	1 ml × 5 tubes

## Storage

Store at -20°C.

## Caution

### Avoid RNase contamination

Please keep the environment of experiment clean. Clean gloves and mask should be worn during the experiment. Centrifuge tubes, tips and other supplies used in the experiment must be RNase-free.

## Protocol

### Reverse Transcription

1. Combine the following components in a RNase-free centrifuge tube.

Components	20 µl Reaction
4× EZscript RT Mix II	5 µl
Total RNA	1 ug (100 ng ~ 2 µg)
or Poly(A)+ RNA	50 ng (10 pg ~ 500 ng)
Nuclease free ddH <sub>2</sub> O	to 20 µl

2. Mix gently by pipetting up and down, then centrifuge the mixture briefly to the bottom of the tube.

3. Perform the reverse transcription reaction at 42°C for 15 minutes and 95°C for 30 seconds.

4. The reverse transcription reaction product can be directly used in PCR applications, but the cDNA is recommended to dilute and mix thoroughly before use (The specific dilution factor depends on the abundance of gene expression. Generally, cDNA is diluted 5 ~ 10 times). If the qPCR experiment is not performed immediately, it is recommended to store at -80°C for long-term storage. Avoid repeated freeze-thaw cycles.