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LiScript™ Fast RT Master Mix

Cat. #: M0038 Size: 200 rxns

Introduction

LiScript™ Fast RT Master Mix is a kit for quick reverse transcription. 5× LiScript™ RT Master Mix contains all the components needed for first strand cDNA synthesis. The efficiency and yield of cDNA firststrand synthesis are higher, and the first strand of cDNA can be synthesized using pg total RNA or mRNA. The reaction can be completed in 15 minutes. This kit is suitable for the high throughout synthesis of first-strand cDNA and subsequent RT-PCR, RT-qPCR, and construction of full-length cDNA libraries.

Package Information

Components	M0038
5× LiScript™ RT Master Mix	400 µl
RNase-Free Water	2× 1 ml

Storage

Store at -20°C

Preparation before the experiment and precautions

1. RNase contamination should be avoided during operation to prevent RNA degradation or cross-contamination in experiments. We suggest that the RNA experiments should be performed in a specialized area with specialized equipment and consumables. The operator should wear a mask and disposable gloves and change gloves frequently.

2. The reaction should be set up on ice to prevent RNA degradation. The enzymes should be returned to -20°C as soon as possible after use to avoid repeated freezing and thawing.

3. For RNA templates with complex secondary structures, it is recommended to incubate the template RNA for 5 minutes at 65°C first, then place it on ice immediately, and centrifuge briefly for further processing.

Protocol

Thaw the template RNA on ice; the kit components should be immediately placed on ice after thawing at room temperature. Each solution should be vortexed to mix, and centrifuge briefly prior to use.

Reverse transcription reaction:

1. Set up the reaction on ice according the following table. :

Reagent	10 µl Reaction
RNA template	x µl (1 pg-0.5 µg)*
5× LiScript™ RT Master Mix	2 µl
RNase-Free Water	Up to 10 µl

Note: 1. If the total RNA is greater than 1 µg, scale up the reaction system. 2. 5× LiScript RT Master Mix contains reverse transcriptase, RNase Inhibitor, Random 6 mers, Oligo(dT) Primer, dNTPs, LiScript RT Buffer *etc*.

2. Vortex to mix well; Briefly centrifuge to collect all the solution to the bottom of the tube.

3. Incubate at 37°C for 15 minutes then incubate at 85°C for 5 minutes.

Note: a. For downstream regular PCR, incubate at 37°C for 30-50 minutes. b. For templates with complex secondary structure, or high GC content, increase the temperature for reverse transcription to 50°C to increase the reverse transcription efficiency.

4. After the reaction is done, briefly centrifuge, then put on ice. For long time storage, please put it in -20°C.

Note: for real-time PCR reaction, the volume of reverse transcription product should NOT exceed the 1/10 volume of the total PCR reaction.

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