WizScript™ cDNA Synthesis Kit

RUO For Research Use Only

REF W2201 (2202)

DESCRIPTION

WizScript™ cDNA Synthesis Kit is a complete system for the efficient synthesis of first strand cDNA from RNA templates. The recombinant Ribonuclease Inhibitor, supplied with the kit effectively protects RNA template from degradation. The kit is also supplied with both oligo dT and random primers. The oligo dT anneals selectively on the poly(A) tail of mRNA. Random primers do not require the presence of poly(A). Therefore, they can be used for transcription of the 5′-end regions of mRNA. Gene-specific primers may also be used with the kit. The first strand of cDNA can be directly used as a template in PCR.

KIT CONTENTS

Contents	W2201	W2202
WizScript™ RTase (200U/μI)	50 μΙ	100 μΙ
10X Reaction Buffer	100 μΙ	200 μΙ
RNase Inhibitor (40U/µI)	25 µl	50 μl
dNTP mix (2.5mM)	100 μΙ	200 μΙ
Oligo dT ₂₀ primer (50pM)	50 μl	100 μΙ
Random hexamer (50pM)	100 μΙ	200 μΙ
DTT (100 mM)	50 μl	100 μΙ
RNase free water	1 ml	1 ml

STORAGE CONDITIONS

Store all components at -20°C in a non-frost-free freezer.

OUALITY CONTROL ANALYSIS:

In accordance with Wizbiosolutions Inc. ISO 13485-certified Quality Management System, each lot WizScript™ cDNA Synthesis Kit (High Capacity) is tested against predetermined specifications to ensure consistent product quality.

PROTOCOL

1. Prepare the following mixture in a microtube.

Reagent	Volume	
Oligo dT20 (Random hexamer)	1 μΙ (2 μΙ)	
dNTP mix (2.5mM)	1 μΙ	
Template RNA *	total RNA : < 5 μg	
	poly(A)+ RNA : < 0.5 μg	
RNase free Water	up to 10 μl	

* Notes: Recommended amounts of RNA template and primers for first-strand cDNA synthesis.

(1) RNAs: total RNA: 10 ng - 5 μg

poly(A)+RNA: 1 ng - 500 ng

(2) Primers: Oligo dT20: 50µM

Random hexamer : 50-100µM Gene-Specific Primer : 15 - 20 pM

2. Keep for 5min. at 65°C & cool immediately on ice.

3. Prepare the reaction mixture by followings

Reagent	Volume
Template RNA & primer mix	10 µl
10X reaction buffer	2 μΙ
RNase Inhibitor (40U/μl)	0.5 µl
RTase (200U/μl)	1 μΙ
DTT (100mM)	1 μΙ
RNase free water	up to 20 µl

- 4. Mix gently
- 5. Incubate the reaction mixture at 42°C/30-60min. (** only random primer is used for 37°C/60 min.)
- Stop the reaction by incubating the tube at 70°C for 10 min. and cooling on ice.
- 7. Synthesized cDNA is immediately used as template for PCR or store at -20°C.

The cDNA can be used immediately, without purification, for end-point or real-time PCR or stored at -20°C for future use.

Notes

- 1. Isolation of poly(A)+ RNA from total RNA is not mandatory, however, doing so may improve the yield and purity of the final product.
- 2. RNA sample must be free of contaminating genomic DNA.
- 3. Unlike the oligo dT priming, which usually requires no optimization, the ratio of a random primer to RNA is critical in terms of the average length of cDNA synthesized in the reaction. Increasing the ratio of random primer/RNA will result in higher yield of shorter (-500 bp) cDNA, whereas decreasing this ratio will produce longer products.
- 4. The synthesized cDNA should be stored at -20°C.

ORDERING INFORMATION

Product	Cat No.	Package
WizScript™ cDNA Synthesis Kit	W2201	50 rxn
	W2202	100 rxn

Technical Support



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