HiSense™ cDNA Synthesis Master Mix

Cat. No. CDS-100, CDS-200, CDS-400

1. Product Information

Introduction

HiSense™ cDNA Synthesis Master Mix is a product formulated in an All-in-one format, allowing for more convenient and rapid synthesis of first-strand cDNA.

The Master Mix contains *M-MLV (Moloney Murine Leukemia Virus)* reverse transcriptase (RTase), ribonuclease inhibitor, dNTPs and an optimized ratio of Oligo (dT)s and random primers.

Primer information

Oligo (dT)s are oligonucleotides that anneal to the 3'-Poly(A) tail of mRNAs. The utility of Oligo (dT) is restricted to mRNA or total RNA templates with 3'-Poly(A) tails. Random Primers anneal at non-specific sites within RNA template(s), they can be used for all forms of RNA as template for cDNA synthesis.

General notes

1) Both poly(A) + mRNA and total RNA can be used for first-strand cDNA synthesis, but poly(A) + mRNA may give higher yields and improved purity of final products.
2) RNA samples must be free of genomic DNA contamination.
3) To remove RNA complementary to the cDNA, add 1 µl (2 U) of E. coli RNase H and incubate at 37°C for 20 mins.
4) Upon completion of the first-strand cDNA synthesis, the cDNA product can be directly applied as a template in a standard PCR/qPCR.

2. Contents and Storage

Materials Provided

Label	CDS-100	CDS-200	CDS-400
All-In-One 5X cDNA Master Mix	200 µl	400 µl	800 µl
Nuclease free water	1 ml	2 ml	4 ml

Storage

Store at -20°C
Check the label on the product for expiration date.

3. Test Protocol

Protocol

1) Thaw RNA templates and the All-In One 5X cDNA Master Mix on ice. Mix solutions gently but thoroughly.
2) Prepare the following reaction mixture in a PCR tube on ice
3) Mix the components well and collect by brief centrifugation. Incubate the mixture in the following reaction conditions.
4) The newly synthesized first-strand cDNA is ready for immediate downstream applications, or for long-term storage at -20°C.

Reaction mixture (for 10µl reaction)

Reaction components	Volume
All-In-One 5X cDNA Master Mix	2 µl
Template RNA*	Variable
Nuclease free water	up to 10 µl
Total volume	10 µl

*The scale of the reverse-transcription reaction can be increased as necessary. Reverse transcription of as much as 500 ng of total RNA is possible with 10 µl of reaction solution.

PCR reaction condition

Steps	Temp(℃)	Time
Primer extension	25	5 min
cDNA synthesis*	42	15 min
Reaction Termination	85	5 sec

*The reverse transcription time can be increased by 15 to 60 minutes or more, depending on the size of the template RNA.