Ver. 1903-00

HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*]

Kit Contents

HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*]		
Cat. No.	**ORTHT100 (100 rxns)**	**ORTHT500 (500 rxns)**
Enzyme Mix [*Hot-Taq*]	0.2 ml	0.2 ml x 5ea
2x Reaction Mix [Hot-Taq] (containing dNTP mix, $MgCl_2$)	1.25 ml x 2ea	1.25 ml x 10ea
Instruction for Use	1ea	1ea

*** Store at -20℃**

Description

HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*] is designed for sensitive amplification of the target gene in one-tube reaction from total transcripts. A reverse transcriptase, HelixCript™ *Thermo* Reverse Transcriptase, and a HelixAmp™ *Hot-Taq* polymerase are supplied as an enzyme mixture.

HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*] : High specific and sensitive RT-PCR

One-step RT-PCR system provide the several advantages.

- Synthesis of cDNA and PCR amplification corresponding to target gene in one-tube reaction
- Obtain the reproductive data in the repetitive experiment(s)
- Can save the time and cost for preparation of RT-PCR
- Amplification of low-copy transcripts by RT-PCR

Application

Detection of target gene transcript from RNA

Semi-quantitative, quantitative analysis of RNA transcription level

Quality control assay data

Functional analysis

The activity for cDNA synthesis and PCR amplification of target gene transcript using HelixCript™ One-Step RT-PCR Kit was evaluated by Limit-of Detection (LOD) assay and long range of gene constitutively-expressed in human total transcripts.

Quality authorized by Yountaek Go

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Protocol

1. **Program the thermal cycler as follows in order to synthesize cDNA using HelixCript™ One-Step RT-PCR Kit with HelixAmp™ *Hot-Taq* .**

Step	Condition		Cycle(s)
cDNA Synthesis		42 ~ 55℃ for 30 ~ 50 min	1
Pre-denaturation		95℃ for 12 ~ 15 min	1
PCR Amplification	Denaturation	95℃ for 20 sec	30 ~ 40
	Annealing	[1)]**AT**℃ for 40 sec	
	Extension	72℃ for 1 min/kb **Collect the fluorescence data**	
Post extension		72℃ for 5 min	1

[1)] **AT,** annealing temperature of primers used

Annealing Temperature = T_m – (6 ~ 8℃)

Where, T_m (Melting Temp.) = [4℃ x (G + C)] + [2℃ x (A + T)]

2. **Add the following components into 0.2 or 0.5 ml micro-tube.**

Components	Volumes
2x Reaction Mix [Hot-Taq]	25 µl
RNA Template (1 ng ~ 5 µg)	X µl
Forward primer (10 pmoles/µl)	2 µl
Reverse primer (10 pmoles/µl)	2 µl
Enzyme Mix [*Hot-Taq*]	2 µl
RNase-free water	to 50 µl

※ RNAs : Total RNA : 10 ng ~ 5 µg, Poly(A)$^+$ RNA : 1 ng ~ 500 ng

3. **Gently mix and immediately centrifuge the reaction mix.**

4. **Perform the one-step RT-PCR.**

Products

Cat. No.	Products	Size
ORTHT100	HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*]	100 rxns
ORTHT500	HelixCript™ One-Step RT-PCR Kit [*Hot-Taq*]	500 rxns

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