

HiSense™ RT-PCR Master Mix

Cat. No. RTPM-1, RTPM-5

1. Product Information

Introduction

HiSense™ RT-PCR Master Mix offer a single-step procedure for performing RT-PCR reactions. Subsequent loading and visualization of RT-PCR products are streamlined since the loading dye are included. The user supplies the primers and DNase-treated template RNA.

This product contains all necessary reagents for PCR amplification to occur in a single reaction tube. Specifically, this One-Step RT-PCR master mix contains special reaction buffer, RTase Inhibitor, dNTPs and DNA-free Taq DNA Polymerase for highly sensitive and specific RT-PCR using any RNA template. The special RT-PCR buffer contains stabilizers and enhancers that optimize the two reactions in a "single step".

The Master Mix offers the end-users an efficient, easy to use and reliable alternative to conventional "two-step" sequential RT-PCR

2. Contents and Storage

Materials Provided

Label	RTPM-1	RTPM-5
2X RT-PCR Master Mix	1 ml	5 ml

Storage

Store at -20°C

Check the label on the product for expiration date.

3. Test Protocol

Reaction mixture (for 20µl reaction)

Reaction components	Volume	
2X RT-PCR Master Mix	10 μl	
Forward primers, (10pmol/μl)*	1 μl	
Reverse primers, (10pmol/μl)*	1 μl	
Template RNA**	2 μl	
Nuclease free water	up to 20 μl	
Total volume	20 μl	

^{*} A final primer concentration of 0.2µM is optimal in most cases but may be individually optimized in a range of 0.1µM to 1.0µM.

PCR reaction condition

Steps & Cycles	Temp(°C)	Time	Cycles
Reverse Transcript*	42	15 min	1
Pre heat	95	5 min	1
Denature	95	30 sec	
Anneal**	60	30 sec	30~40
Extend***	72	1 min	
Final extension	72	5 min	1

^{*} The reverse transcription time can be increased by 30 to 60 minutes or more, depending on the size of the template RNA.

^{**} Use 10 pg-100 ng of total RNA as template.

^{**}Optimal annealing temperature depends on the melting temperature of the primers.

^{***} Generally, 1 min/kb and higher than 3 kb, set it to 1.5 to 2.0 min/kb.