

Imagen ® TaqMan multiplex RT-qPCR Premix

Cat. No

Description

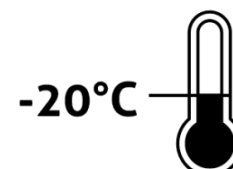
Imagen ® TaqMan multiplex RT-qPCR Premix is a ready-to-use and to amplify up to four detect targets at the same time. It is 2x concentrated premix including a novel Hotstart Taq DNA polymerase 、Fast Reverse transcriptases 、optimized reaction buffer and nucleotides with combines accurate using sequence-specific fluorogenic probe chemistries including hydrolysis probes(ex. TaqMan 、Molecular beacon,etc...) which provides good specificity and amplification efficiency for real-time RT-PCR.

Contents

The **Imagen ® TaqMan multiplex RT-qPCR Premix** is supplied as a ready-to-use 2x reaction mix. The formulation contains, Hotstart Taq DNA polymerase, Fast Reverse transcriptases, dNTPs, MgCl₂, reaction enhancers, and stabilizers.

Storage

- ✓ -20 °C
- ✓ Protected from light
- ✓ Avoid repeated freezing and throwing



Reaction Mix Thawing and Handling

Imagen ® TaqMan multiplex RT-qPCR Premix is delivered in a 2x ready-to-use format. To use the mix, thaw the vial on ice to 4 °C.

Please completely mix the vial and briefly centrifuge to ensure all components are at the bottom of the tube. Store on ice protected from light until ready to use. If using automated liquid handling, let sit at ambient temperature for 10 min to further reduce the viscosity.

Application

- ✓ Quantitative 1-step real-time RT-PCR
- ✓ Gene expression analysis
- ✓ Low copy gene detection

Prepare the qPCR Reaction Mix

1. Mix the **Imagen ® TaqMan multiplex RT-qPCR Premix** thoroughly but gently until it's completely
2. homogenous.
Prepare the qPCR Reaction Mix for the number of reactions required as shown in table below and plus 10% overage.

Reagent	Volume (ul)	Final conc.
Imagen ® TaqMan multiplex RT-qPCR Premix	12.5	1x
Forward Primer(10 uM)	0.75	300 - 600 nM
Reverse Primer(10 uM)	0.75	300 - 600 nM
Fluorogenic Probe(10 uM)	0.5	200 nM
RNA Template or NTC(Nuclease-free water)	2	100 ng - 10 pg
Nuclease-free water	8.5	-
Final volume	25	-

3. Vortex the tube to mix the contents thoroughly, then centrifuge briefly to collect the contents at the bottom of the tube. (*Use good pipetting practice to ensure assay precision and accuracy of dispensing.)

4. Add DNA (and nuclease-free water, if needed) to the PCR tubes or wells containing the reaction mix, seal tubes or wells with flat caps or optically transparent film, and gently vortex to ensure thorough mixing of the reaction components.
5. Program the thermal cycling protocol on the real-time PCR instrument.

Step	Temp. °C	Time	Cycles
Reverse Transcription	48°C	15 min	1
DNA polymerase activation and template denaturation Amplification	95°C	10 min	1
Amplification	95°C	10 sec	35-40
	58 - 62°C	≥ 30 sec © Data acquisition	

6. Load the PCR tubes or plates onto the real-time PCR instrument and start the qPCR run program.
7. When thermal cycling is complete, perform data according to the instructions in the instrument-specific software.