Imagen ® TaqMan qPCR Premix

Cat. No

Description

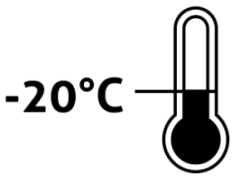
Imagen ® TaqMan qPCR Premix is a ready-to-use, 2X concentrated premix that contains all reagents (except primers, probe and template.) need for running real-time PCR. Real-time PCR is performed by addition of various probes (ex. Taq Man, Molecular Beacon,etc…). The ROX is optimal for instruments from Applied Biosystems (models 7000,7300, 7700, 7900HT, StepOne™, and StepOnePlus™, but not Applied Biosystems 7500 Real-Time PCR Systems、 Smart Cycler® or LightCycler® real time instruments.) The Passive Reference (ROX) does not participate in the PCR amplification.This product combines the high performance Hotstart Taq with a buffer which provides good specificity, amplification efficiency for real-time PCR.

Contents

The **Imagen ® TaqMan qPCR Premix** is supplied as a ready-to-use 2x reaction mix. The formulation contains, Hotstart Taq DNA polymerase, dNTPs, $MgCl_2$, ROX Reference Dye, reaction enhancers, and stabilizers.

Storage

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



Reaction Mix Thawing and Handling

Imagen ® TaqMan 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

- ✓ TaqMan qPCR based on Specific Probes.
- ✓ Detection and Quantification of DNA targets.
- ✓ High Throughput Applications.

Prepare the qPCR Reaction Mix

1. Mix the **Imagen ® TaqMan qPCR Premix** thoroughly but gently until it's completely homogenous.
2. 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 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
DNA Template	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. ℃	Time	Cycles
DNA polymerase activation and template denaturation Amplification		95℃	10 min	1
Amplification	Template denaturation	95℃	20 sec	35-40
	Annealing / Extension and plate read	58 - 62℃	60 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.