Product Use Limitation & Warranty

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OneStep RT-PCR Kit

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Cat. No. FYT503-50P FYT503-100P

Deoxy⁺ OneStep RT-PCR Kit

Concentration: 2X

Storage: - 20 °C

Description

The Deoxy⁺ OneStep RT-PCR Kit is a ready-to-use master mix which eliminates the need for optimization of reaction and cycling conditions for one-step RT-PCR. The reaction can be prepared by simply adding template RNA and primers to the master mix. The use of Yeastern's RealStart DNA polymerase and Deoxy⁺ HiSpec RT enables reliable real-time RT-PCR quantification on any real-time PCR machines. Since it is a one-tube reaction, the procedure makes high-throughput analysis possible. After reverse transcription, reactions are heated to 95°C for 10 minutes to inactivate the reverse transcriptase and simultaneously activate RealStart Taq DNA polymerase. This one-step formulation eliminates any nonspecific amplification products such as primer-dimers and reduces background smear, ensuring highly sensitive and reproducible RT-PCR.

Content

2× OneStep RT-PCR Premix containing:

- RealStart Taq DNA polymerase
 Deoxy+ HiSpec Reverse Transcriptase
- dATP : dCTP : dGTP : dTTP mix
- dair, dcir, dcir, diir mix
- 5 mM MgCl₂

Procedure

A. Preparing the master mix on ice

| | Component | Volume | Final conc. |
|--|----------------------------|----------|-------------|
| | 2× OneStep RT-PCR premix | 12.5 µl | 1× |
| | Forward primers (5-10 µM) | variable | 0.6-1.0 μM |
| | Reversed primers (5-10 µM) | variable | 0.6-1.0 μM |
| | ddH₂O | variable | |
| | | | |

Total volume 23 µl







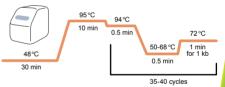
B. Mix by pipetting

C. Add 2 µl of the RNA

D. Mix carefully by pipetting



E. Program your instrument





F. PCR tubes are kept on ice until the thermal cycle has

reached 48 °C. Then place the PCR tubes in the thermal cycle and start the RT-PCR program.



OneStep RT-PCR Premix Performance Test

High sensitivity of cDNA amplification by OneStep RT-PCR is confirmed by using 10 copies viral RNA as the template (amplified fragment: 500 bp).