2x RT-Master mix

Cat No: ORT01
Size: 50 rxn / 500ul

Description :

The 2X RT master mix is designed for convenient and efficient cDNA synthesis. The 2X pre-mixed reagent containing RTase, Random 6mers, oligo dT ,dNTP mixture and reaction buffer. The reverse transcription reaction just add RNA and water. The protocol is easy and the reaction can be completed in short time.

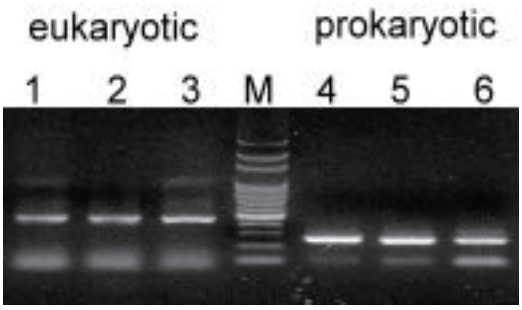
Storage conditions: long time at -20°C

2X RT Master Mix :

Reverse transcriptase
2X reaction buffer
dNTP mix
oligo dT primers and random hexamers.
stabilizer

Application

cDNA synthesis
PCR screening
Real-time PCR



Lane 1-3 is soybean gene βactine, Lane 4-6 is Ecoli gene
1: M-MLV RTase + oligo dT
2: 2X master mix
3: M-MLV RTase + specific gene primer
M:100 bp DNA ladder
4: M-MLV RTase + random 6mer
5: 2X master mix
6: M-MLV RTase + specific gene primer

Protocol

1.Thaw the 2x RT master mix at room temperature. Vortex the 2x RT master mix and then spin it briefly in a micro centrifuge to collect the material in the bottom of the tube.

2. Incubate the RNA 0.1-5 ug (optional add 2ul 5X RNA Protector) in RNase free ddH2O to final volume 10 ul and stand at 65°C for 15 min, and then keep on ice.

3. Prepare one of the following reaction mixes on ice:

Component	Volume
2xRT master mix	10 ul
heat treated RNA	0.1-5 ug/ 10 ul
Total	20 ul

4. Incubate the mixture at 37-42°C during 30-120 min. The time of reaction depends on the length of cDNA, 30 min is for cDNA in range of 500 bp, 120 min is for cDNA more then 1.5 kb. The temperature of the reaction depends on the structural features of RNA.
Use increased temperature (up to 50°C) for the highly structured RNA when treated 37-42°C for 10mins later.

5. Heat the mixture 5 min at 80°C to inactivate the RTase.

6. Use the mixture for PCR or for other application.

For your PCR-Reaction you need 1-10 µl of your RT product.

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