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Easy™ cDNA Synthesis Kit*(New)*

Cat A101161 (50 Reactions)
Store at -20°C

Contents

Component	Volume
Buffer-Mix (2x)	500 µL
Enzyme Mix	100 µL
DEPC-treated water	500 µL
cDNA Con. Primer Mix(B2M)	50µL

Description

Easy cDNA Synthesis kit contains all necessary components for conversion of total RNA or mRNA to ***the single stranded cDNA***. The 2X Buffer mix solutions contains, RTbuffer, 1mMdNTP mixture, 8mM MgCl2,Oligo d(t)16, Randomhexamer and stabilizer. Enzyme mix contains thermostable H-minus MMLV, RNase Inhibitor and stabilizer.

Features

- Easy protocol& Minimum pipetting steps
- RNase minus MMLV enzyme
- Long mRNA synthesis
- High temperature reaction to destabilize RNA secondary structures

General Reaction Protocol (first strand cDNA synthesis):

1- Mix the template RNA (total RNA orPoly(A)mRNA) and other kit components in RNase-free tube as below table.

Template RNA	1 ng~5 µg	XµL
Buffer-Mix (2x)		10
Enzyme Mix		2
DEPC-treated water		**Up to 20 µL**

2- Mix the above mixture by quick vortex.
3- Incubate 10 min at 25 °C.
4. Incubate 60 min at 47°C.
5. Stop the reaction by heating at 85°Cfor 5 minutes. Chill on the ice or at 4°C.

Note:To perform PCR, you can add the finished RT reaction up to 1/5 of the final PCR volume.

cDNAControl PCR Reaction

1- Prepare a reaction mix according to the table.
2- For negative tube use 2µl of PCR grade water. The final volume in each PCR reaction tubes is 20µl.

Component	Vol (µL)
Taq 2X Premix	10
cDNA Control primer mix	1
cDNA	2
PCR Grade Water	7
Final volume	**20 µL**

Note: It is recommended that all of the PCR components be premixed in a sufficient quantity for daily needs and then dispensed into the individual reaction tubes.

Amplification protocol

Cycle	Time	Temp °C
1	4 Min	95
35	30 Sec	94
	30 Sec	57
	30 Sec	72
1	5 Min	72

Agarose gel Electrophoresis

Run the total 5-7 µL of PCR products alongside 3µL DNA marker on a 2% agarose gel containing Green viewer DNA safe stain.
The B2M primers amplify a band of approximately 230bp from human, mouse and rat *B2M* cDNA.

Disclaimers and Addresses

This product is for research use only and should only be used by trained professionals.

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