Lolina A/S

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Product Information

Product name	Lolina® V Universal Multiplex One Step RT-qPCR Probe Kit
Cat.No.	NaM602006
Size	100 T / 500 T / 1000 T / 10000 T
Storage and shipping	1. The product is shipped with ice pack. 2. The product can be stored at -15℃ ~ -25℃ for 18 months.
Application equipment	**ABI** 5700, 7000, 7300, 7700, 7900HT Fast, StepOne™, StepOne Plus™, ABI 7500, 7500 Fast, ViiA™7, QuantStudio ™ 3 and 5, QuantStudio™ 6,7, 12k Flex **Stratagene** MX3000P™, MX3005P™, MX4000P™ **Bio-Rad** CFX96™, CFX384™, iCycler iQ™, iQ™5, MyiQ™, MiniOpticon™, Opticon®, Opticon® 2, Chromo4™ **Eppendorf** Mastercycler® ep realplex, realplex 2 s **Qiagen** Corbett Rotor-Gene® Q, Rotor-Gene® 3000, Rotor-Gene® 6000 **Roche** Applied Science LightCycler® 480, LightCycler® 2.0; Lightcycler® 96 **Thermo Scientific** PikoReal Cycler; **Cepheid** SmartCycler®; **Illumina** Eco qPCR

Product description

Lolina® V Universal Multiplex One Step RT-qPCR Probe Kit is a kit for performing multiplex quantitative PCR reactions using RNA as a template. During the experiment, reverse transcription and quantitative PCR were performed in the same reaction tube, which simplified the experimental operation and reduced the risk of contamination.

This kit uses heat-stable Lolina® V Reverse Transcriptase to efficiently synthesize first-strand cDNA and Lolina® HotStart Taq DNA Polymerase for quantitative amplification. This kit mainly contains optimized MP Buffer, Enzymes Mix, etc. The buffer already contains Mg^{2+} and dNTPs, etc., and has added factors that effectively inhibit non-specific PCR amplification and factors that improve the

amplification efficiency of multiplex qPCR reactions, which can Perform up to four-plex reactions while ensuring primer amplification efficiency.

Components

Compont No.	Name	Size			
		100T	500T	1000T	10000T
A	2×MP Buffer	1.25 mL	6.25 mL	12.5 mL	125 mL
B	Enzyme Mix	100μL	500 μL	1 mL	10 mL

[Note]:

a) 2×MP Buffer are the abbreviation for Multiplex One Step RT-qPCR Probe Buffer.

b) Enzyme Mix mainly contains heat-resistant V Reverse Transcriptase and HotStart Taq DNA Polymerase.

Operate

Reaction System

Components	Volume μL	Final concentration
2×MP Buffer	12.5	1 ×
Enzyme Mix	1	
Primer mix (10 μM)	1 each	0.4 μM
Probe mix (10 μM)	0.5 each	0.2 μM
Template	1 - 10	-
ddH_20	Up to 25	-

[Note]: Be sure to mix thoroughly before use, avoiding excessive bubbles caused by vigorous shaking.

a) Primer concentration: Primer Mix contains multiple pairs of primers and can be adjusted between 0. 1-1.0 μM according to the situation.

b) Probe concentration: Probe Mix contains multiple probes with different fluorescence signals, and the concentration of each probe can be adjusted between 50-300 nM according to specific conditions.

c) Template dilution: The sensitivity of qPCR is extremely high. It is recommended to dilute the template and control the Ct value between 20-35.

d) Reaction system: 25-50 μL is recommended to ensure the effectiveness and reproducibility of target gene amplification.

e) System preparation: Please prepare in a clean workbench, and use pipette tips and reaction tubes without nuclease residues; it is recommended to use pipette tips with filter elements; avoid cross-contamination and aerosol contamination.

Reference reaction program

Cycle step	Temp.	Time	Cycles
Reverse transcription	50 °C [a)]	10 min	1
Initial denaturation	95 °C	5 min	1
Amplification reaction	95 °C	15 sec	45
	60 °C[b)]	30 sec [c)]	

[Note]:

a) Reverse transcription: 42℃ or 50℃ can be used.

b) Amplification reaction: The amplification reaction temperature is adjusted according to the designed primer Tm value.

c) Fluorescence signal collection: Different qPCR detection instruments require different fluorescence signal collection times. Please set according to the minimum time limit.

Notes

1. For your safety and health, please wear lab coats and disposable gloves for operation.

2. Please use RNase free consumables.

3. This product is for research use **ONLY**!