

Hieff Unicon™ V Universal Multiplex One Step RT-qPCR Probe Kit

Product Information

Product Name	Cat#	Size
Hieff Unicon™ V Universal One Step RT-qPCR Probe Kit	11213ES60	100 T
	11213ES76	500 T
	11213ES80	1000 T
	11213ES92	10000 T

Product Description

Hifair™ V Multiplex One-Step RT-qPCR Probe Kit is for multiple quantitative PCR reactions with RNA as a template. During the experiment, the reverse transcription and quantitative PCR were carried out in the same reaction tube, simplified experimental operation and reduced the risk of pollution.

The kit uses heat-resistant Hifair™ V Reverse Transcriptase to efficiently synthesize the first chain cDNA, and use Unicon™ Hotstart Taq DNA Polymerase to quantify quantitative amplification. The kit mainly includes an optimized MP Buffer, Enzymes Mix, etc., which already contains Mg²⁺ and DNTP, etc., and has added factors that effectively inhibit non-specific PCR amplification factors and enhance multiple qPCR reactive amplification efficiency, which can be guaranteed at the same time of primer amplification, up to four reactions.

Product Components

Component Name	Components	Cat#/Size			
		11213ES60 (100 T)	11213ES76 (500 T)	11213ES80 (1000 T)	11213ES92 (10000 T)
11213-A	2×HU™ MP Buffer	1.25 mL	6.25 mL	12.5 mL	125 mL
11213-B	HU™ Enzyme Mix	100 µL	500 µL	1 mL	10 mL

1) 2×HU™ MP Buffer is short of Multiplex One Step RT-qPCR Probe Buffer.

2) HU™ Enzyme Mix mainly includes heat-resistant Hifair™ V Reverse Transcriptase and UNICON™ HotStart Taq DNA Polymerase.

Applicable Models

ABI Series: ABI 5700, 7000, 7300, 7700, 7900HT Fast, StepOne, StepOne Plus; 7500, 7500 Fast, ViiA7, QuantStudio 3 and 5, QuantStudio 6, 7, 12k Flex;

Bio-Rad Series: Bio-Rad CFX96, CFX384, iCycler iQ, iQ5, MyiQ, MiniOpticon, Opticon, Opticon 2, Chromo4;

Roche Series: Roche Applied Science LightCycler 480, LightCycler 2.0; Lightcycler 96;

Others: Stratagene MX3000P, MX3005P, MX4000P;

Eppendorf Mastercycler ep realplex, realplex 2 s;

Qiagen Corbett Rotor-Gene Q, Rotor-Gene 3000, Rotor-Gene 6000;

Thermo Scientific PikoReal Cycler; **Cepheid** SmartCycler; **Illumina** Eco qPCR.

Shipping and Storage

The product is shipped with ice packs and can be stored at -20°C for one year. Please avoid repeated freeze-thaw. It is recommended to save.

Cautions

- 1) Please use the RNase free consumables during the experiment;
- 2) For your safety and health, please wear lab coats and disposable gloves for operation.

Reaction System(Take 25 μ L for example)

Components	Volume	Final Concentration
2 \times HU TM MP Buffer	12.5 μ L	1 \times
HU TM Enzyme Mix	1 μ L	-
Primer Mix (10 μ mol/L)	1 μ L each	0.4 μ mol/L
Probe Mix (10 μ mol/L)	0.5 μ L each	0.2 μ mol/L
Sample RNA	1-10 μ l	-
RNase Free H ₂ O	to 25 μ L	-

[Notes]: Be sure to mix well before use to avoid excessive air bubbles caused by vigorous shaking.

- 1) Primer Concentration:** Primer Mix contains multiple pairs of primers, usually the final concentration of each primer is 0.25 μ mol/L, and can also be adjusted between 0.1-1.0 μ mol/L according to the situation;
- 2) Probe Concentration:** Probe Mix contains multiple probes with different fluorescent signals, and the concentration of each probe can be adjusted between 50-300 nmol/L according to the specific situation;
- 3) Template Dilution:** The sensitivity of qPCR is extremely high. It is recommended to dilute the template and control the Ct value between 20-35;
- 4) Reaction System:** 20 μ L to 50 μ L is recommended to ensure the validity and repeatability of target gene amplification;
- 5) System Preparation:** Please configure in the ultra-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;

Standard Amplification Procedure

Stage	Temperature	Time	Cycles
Reverse Transcription	50°C ^a	10 min	1
Pre-denaturation	95°C	5 min	1
Amplification Reaction	95°C	15 sec	45
	60°C ^b	30 sec ^c	

[Notes]:

- a) Reverse Transcription:** 42°C or 50°C;
- b) Amplification Reaction:** The temperature of the amplification reaction is adjusted according to the T_m value of the designed primers;
- c) Fluorescence Signal Collection:** Different qPCR detection instruments require different fluorescence signal collection times, please set according to the shortest time limit;