

## Hieff Unicon™ Universal TaqMan multiplex qPCR master mix

### Product Information

Product Name	Cat#	Size
Hieff Unicon™ TaqMan Multiplex qPCR Master Mix (UDG plus)	11211ES03	1 mL
	11211ES08	5×1 mL
	11211ES09	5 mL
	11211ES20	20 mL
	11211ES60	100×1 mL
	11211ES61	100 mL

### Product Description

This product is a 2× Mix pre-mixed reagent that enables up to four fluorescent quantitative PCR reactions in a single reaction well. This product contains the genetically modified antibody method to hot-start Taq enzyme, greatly improving the amplification sensitivity and specificity. At the same time, this product has deeply optimized the multi-reaction buffer, which can improve the amplification efficiency of the reaction and promote the effective amplification of low-concentration templates. This product can be used for genotyping and multiplex quantitative analysis.

### Product Components

Component Number	Component	Cat#/Size					
		11211ES03 (1 mL)	11211ES08 (5 × 1 mL)	11211ES09 (5 mL)	11211ES20 (20 mL)	11211ES60 (100×1 mL)	11211ES61 (100 mL)
11211	2× Hieff Unicon™ Universal TaqMan multiplex qPCR master mix	1 mL	5 × 1 mL	5 mL	20 mL	100 × 1 mL	100 mL

### Shipping and Storage

The product is shipped with ice pack and can be stored at -20°C for 1 year.

### Reaction System

Components	Volume (μL)	Final Concentration
2× Hieff Unicon™ Universal TaqMan multiplex qPCR master mix	12.5	1×
Primer mix (10 μmol/L)	x	0.1-0.5 μmol/L
Probe mix (10 μmol/L)	x	50-250 nmol/L
Rox reference dye	0.4	1×
Template DNA/cDNA	1-10	-
ddH <sub>2</sub> O	up to 25	-

[Note]: Mix thoroughly before use to avoid excessive bubbles from vigorous shaking.

- Primer concentration:** Primer Mix contains multiple pairs of primers, usually each primer at a final concentration of 0.2 μmol/L and can also be adjusted between 0.1 and 0.5 μmol/L as appropriate.
- Probe concentration:** Probe Mix contains multiple probes with different fluorescence signals, and the concentration of

each probe can be adjusted between 50 and 250 nmol/L according to specific situation.

- c) **Rox dye reference:** It is used on Real Time PCR amplification instrument such as Applied Biosystems to correct the error of fluorescence signal generated between wells; this product does not contain Rox dye reference. Cas#10200 is recommended if needed.
- d) **Template dilution:** qPCR is highly sensitive, and it is recommended to dilute the template for use. If the template is a cDNA stock solution, the template volume should not exceed 1/10 of the total volume.
- e) **Reaction system:** 25  $\mu$ L, 30  $\mu$ L or 50  $\mu$ L is recommended to ensure the effectiveness and repeatability of target gene amplification.
- f) **System preparation:** Please prepare in the super clean bench, and use the tips and reaction tubes without nuclease residue; it is recommended to use the tips with filter cartridges. Avoid cross contamination and aerosol contamination.

## Reference Program

Cycle step	Temp.	Time	Cycles
Initial denaturation	95°C	5 min	1
Denaturation	95°C	15 s	45
Annealing/Extension	60°C	30 s	

[Note]: a) **Annealing/Extension:** The temperature and time can be appropriately adjusted according to the designed primer  $T_m$  value.

b) **Fluorescence signal acquisition:** The fluorescence signal acquisition time required for different qPCR detection instruments is different, please set according to the minimum time limit. The time of several common instruments is set as follows:

20 sec: Applied Biosystems 7700, 7900HT, 7500 Fast

31 sec: Applied Biosystems 7300

32 sec: Applied Biosystems 7500

## Cautions

### About Operation

1. Please ensure that the product is completely thawed and thoroughly mixed before use, briefly centrifuge to collect to the bottom of the tube, and place on ice for later use.
2. To avoid cross-contamination between samples and aerosol contamination, it is recommended to prepare the reaction system in a super clean bench.
3. Avoid repeated freezing and thawing of the product during use.
4. For your safety and health, please wear lab coats and disposable gloves for operation.

### Experimental Method

- a) Primer design
  1. Primers must be specific.
  2. The  $T_m$  value of the primers must be 58-60°C, and the  $T_m$  difference of each primer group should be controlled within 1-2°C.
  3. The length of primers is generally controlled between 15-30 bp.
- b) Probe Design
  1. The  $T_m$  value of TaqMan probes is approximately 10°C higher (68-70°C) than the corresponding primers.
  2. Inhibit dimerization between TaqMan probes and primers.