

HIFI Multiplex RT-PCR Master Mix

Instruction for Use

【Product Name】

HIFI Multiplex RT-PCR Master Mix

【Cat. No./Spec.】

K006-A (100 rxns); K007-6 (1000 rxns); K006-C (5000 rxns)

【Product Description】

HIFI Multiplex RT-PCR Master Mix is RT-PCR premix for high-throughput sequencing. It contains various components required for RT-PCR reactions (except primers and templates). Through the reverse transcription step, both RNA and DNA target sequences can be amplified. Using high-performance high-fidelity DNA polymerase, the PCR product is blunt-ended. The amplification reagent has undergone strict quality control and functional verification to ensure the stability and repeatability of the reagent to the greatest extent.

【Components】

Component	K006-A (100 rxns)	K006-B (1000 rxns)	K006-C (5000 rxns)
HIFI Multiplex RT-PCR Master Mix	1 mL × 1	10 mL × 1	10 mL × 5

【Storage Condition & Shelf Life】

Store unopened at -15°C to -25°C until the expiration date on the label. After opening, the master mix may be stored at -15°C to -25°C until the expiration date on the label, or at 4°C for up to 30 days.

【Application】

This product is suitable for multiplex amplification of DNA/RNA templates.

【Protocol】

Note: Before setting up the PCR reactions, prepare a primer mix with $5\ \mu\text{M}$ of each primer.

1. Prepare the PCR Reaction Mix

1.1 Allow all reagents to thaw completely. Mix gently by inverting the tube. Spin briefly. Put all reagents on ice.

1.2 The reaction system is formulated according to the following table:

Reagents	Volume	Final Conc.
HIFI Multiplex RT-PCR Master Mix (2X)	10 μL	1X
Primer mix (5 μM each)	1 μL	250 nM each primer ^[1]
Template DNA/RNA	X μL	-
Nuclease-free water	To 20 μL	n/a

[1] The amount of primers varies greatly depending on the application scenario, please set the primer concentration gradient according to the experiment to find the optimal concentration.

2. Put the tube into PCR machine for amplification according to the following procedure:

Step	Time	Temp. ($^{\circ}\text{C}$)
Hold	20 min	55
Hold	1 min	95
20~32 Cycles	15 sec	98
	30 sec	60
	30 sec	72
Hold	2 min	72
Hold	∞	4

【Notes】

- High-quality multiplex primers are one of the key factors for effective amplification. Minimize cross-complementarity between primers and each other.
- DNA polymerase uses fast high-fidelity enzymes. Adjust the amplification time according to the actual amplification product length.
- This product adopts well-tolerated reverse transcriptase and DNA polymerase, which can detect large or crude samples.

【Library Preparation Module】

GDSBio offers the following DNA and RNA library construction modules that can be used in combination for high-quality library preparation:

Module	Product Name	Cat. No./Spec.
cDNA First Strand Synthesis	GDS RNA First Strand Synthesis Module	K020-A/24 rxns K020-B/96 rxns
Directional cDNA Second Strand Synthesis	GDS Directional RNA Second Strand Synthesis Module	K021-A/24 rxns K021-B/96 rxns
Non-Directional cDNA Second Strand Synthesis	GDS Non-Directional RNA Second Strand Synthesis Module	K022-A/20 rxns K022-B/100 rxns
Fragmentation & End Repair	GDS Fragmentation & End Prep Module	K023-A/24 rxns K023-B/96 rxns
Fragmentation	GDS dsDNA Fragmentase	K024-A/50 rxns K024-B/250 rxns
End Repair/dA-Tailing	GDS End Preparation Module	K025-A/24 rxns K025-B/96 rxns
Adapter Ligation	GDS Ligation Module	K026-A/24 rxns K026-B/96 rxns
Amplification	HIFI Library PCR Master Mix	K007-A/40 rxns K007-B/400 rxns K007-C/2000 rxns
Cleanup/Size Selection	GDSPure DNA Selection Magbeads	NC1011/5 mL NC1012/60 mL NC1013/450 mL

This product is for research use only.