



## Super PCR mastermix

2X super PCR MasterMix (Dye Plus)	1 ml
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### **For research use only**

Cat No : YT1553

Size: 1 ml

Store at  $-20^{\circ}\text{C}$ 

### Introduction

2x Super PCR mastermix is a mix containing taqplus enzyme. TaqPlus enzyme is a mixture of taq DNA polymerase and PFU DNA polymerase. PFU is a DNA polymerase containing 3'→5' exonuclease activity. Taq Plus DNA polymerase has stronger amplification performance and higher sensitivity. Super PCR MasterMix contains everything required for PCR, except primers and template, thereby this ready to use mastermix easing PCR setup and improving reproducibility.

2X super PCR MasterMix contains everything required for PCR, except primers and template, thereby easing PCR setup and improving reproducibility. It can amplify up to **10 kb** from human genomic DNA or up to **15 kb** from  $\lambda$  DNA. Protective agents in the 2X super PCR MAsterMix enable the resistance to repeated freeze-thaw cycles.

Dye mixed to 2X super PCR MasterMix enable direct loading PCR products onto agarose gels. The obtained PCR products contain A at the 3'ends and can be directly cloned into T-Vectors.

**Composition :** Taq DNA polymerase , Buffer , dNTPs , 3mM MgCl<sub>2</sub> , dye and stabilizer

### Protocol

General reaction Mixture for PCR	50 $\mu$ l reaction	25 $\mu$ l reaction
ddH <sub>2</sub> O	To 50 $\mu$ l	To 25 $\mu$ l
Super PCR mastermix 2X	25 $\mu$ l	12.5 $\mu$ l
Template DNA*	Optional	Optional
Primer1 (10pM)	1 $\mu$ l	0.5 $\mu$ l
Primer 2 (10pM)	1 $\mu$ l	0.5 $\mu$ l

### \*Recommended amount of DNA template for a 50 $\mu$ l reaction system is as follow:

Human genomic DNA	0.1~1 $\mu$ g
Bacterial Genomic DNA	10~100ng
$\lambda$ DNA	0.5~5ng
Plasmid DNA	0.1~10ng

### Thermocycling Condition for a routine PCR

94°C	5 min (pre-denaturation)	
94°C	30 sec	} 35 Cycles
55°C *	30 sec	
72°C	60 sec/kb	
72°C	7 min (final extention)	
4°C	Hold	

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**\*The optional annealing temperature should be 1-2°C lower than the  $T_m$  of primers used.**

**Primers Designing Notes**

1. Choose C or G as the last base of the 3' end of the primer;
2. Avoid continuous mismatching at the last 8 bases of the 3' end of the primer;
3. Avoid hairpin structure at the 3' end of the primer;
4.  $T_m$  of the primers should be within the range of 55°C~65°C;
5. Additional sequence should not be included when calculating  $T_m$  of the primers;
6. GC content of the primers should be within the range of 40%~60%;
7.  $T_m$  and GC content of forward and reverse primes should be as similar as possible.