

Catalog # P131

Ŵ Vazyme

Version 5.1

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#### Introduction

Green Taq Mix is a ready-to-use 2× PCR mix that contains Taq DNA Polymerase, dNTPs, and an optimized buffer system. The amplification can start only with the addition of primer and template, thereby reduces pipetting steps and improves throughput, specificity, and reproducibility. Protective reagents in the Green Taq Mix enable its resistance to repeated freeze-thaw cycles. The mix contains a green loading dye which enable direct loading PCR products onto agarose gels for electrophoresis. The obtained PCR products are compatible with ClonExpress II One Step Cloning Kit series (Vazyme, Cat.No. #C112, #C113). The PCR products contain A at the 3'-end and can be directly cloned into T-Vectors.

# **Package Information**

Components	P131-01	P131-02	P131-03	
Green Taq Mix	5 ml	15 ml	50 ml	
Components	P131-w1	P131-w2	P131-w3	
Green Taq Mix	5 ml	15 ml	50 ml	
ddH <sub>2</sub> O	5 ml	15 ml	50 ml	

#### Storage

Store at -20°C for 1 year, or store at -4°C for 2 months.

# **Application**

Conventional PCR Colony PCR RT-PCR Genotyping Amplification of fragments with high AT-content.

# Protocol

#### 1. General reaction mixture for PCR:

ddH <sub>2</sub> O	to 50 µl			
Grean Taq Mix	25 µl			
Template DNA*	Optional			
Primer 1 (10 µM)	2 µl			
Primer 2 (10 µM)	2 µl			
*The recommended amount of DNA template for a 50 µl reaction system is as follows:				
Human Genomic DNA	10 - 200 ng			
Bacterial Genomic DNA	10 - 100 ng			
λDNA	0.5 - 5 ng			
Plasmid DNA	0.1 - 10 pg			



#### 2. Thermocycling conditions for a routine PCR:

94°C	5 min (Pre-denaturation)	
94℃	30 sec	)
58℃*	30 sec	> 30 - 35 cycles
72℃	60 sec / kb	)
72°C	7 min (Final extension)	
4℃	Hold	

\*The optimal annealing temperature should be  $1^{\circ}$ C -  $2^{\circ}$ C lower than the T<sub>m</sub> of the primers used.

#### **Notes for Electrophoresis**

The PCR product can be directly loading to a 1% agrose gel for electrophoresis. In the result, the band of blue dye is approximately at 4 kb, while yellow dye is at < 50 bp.

# **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 Tm 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.



