



# Experiment Process

## Reaction System

ddH <sub>2</sub> O	To 50 µl
2 × Taq Master Mix	25 µl
Primer 1 (10 µM)	2 µl
Primer 2 (10 µM)	2 µl
Template DNA*	x µl

\*Optimal reaction concentration varies in different templates. In a 50 µl system, the recommended template usage is as follows:

Animal & Plant Genomic DNA	0.1 - 1 µg
<i>E. coli</i> Genomic DNA	10 - 100 ng
cDNA	1 - 5 µl (≤1/10 of the total volume of PCR system)
Plasmid DNA	0.1 - 10 ng
λDNA	0.5 - 10 ng

## Reaction Program

95°C	3 min (Initial Denaturation) <sup>a</sup>	} 30 - 35 cycles
95°C	15 sec	
60°C <sup>b</sup>	15 sec	
72°C	60 sec/kb	
72°C	5 min (Final Extension)	

- a. The condition of initial denaturation is applicable for most amplification reactions and can be adjusted according to the complexity of the template structure. If the template structure is complex, the initial denaturation time can be extended to 5 - 10 min to improve its effect.
- b. The annealing temperature needs to be adjusted according to the T<sub>m</sub> value of the primer, generally set to be 3 ~ 5°C lower than the T<sub>m</sub> value of the primer; For complex templates, it is necessary to adjust the annealing temperature and extend the extension time to achieve efficient amplification.

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