

**Features**

Specific features of PCR enzymes are listed here in Table 1.

**Table 1.** PCR enzymes features and specifications

<div style="text-align: center;"><b>Kit Name</b></div> <div style="text-align: left;"><b>Feature</b></div>	<b>Si-Taq Polymerase****</b>	<b>Hy-Taq Polymerase****</b>	<b>5t-pfu Polymerase**</b>	<b>Hy-Fidelity Pfu Polymerase</b>
<b>Fidelity***</b>	1	2	18	54
<b>Expression rate</b>	1-2 kb/min	6 kb/min	0.5 kb/min	2-4 kb/min
<b>Amplification of genomic DNA fragment up</b>	3 kb	4 kb	6 kb	15 kb
<b>Amplification of plasmid DNA fragment up</b>	—	—	10 kb	20 kb
<b>Hot-start</b>	No	No	No*	Yes
<b>Applications</b>				
<b>Short fragment PCR</b>	✓	✓	✓	✓
<b>High throughput PCR</b>	x	✓	x	x
<b>Colony PCR</b>	x	✓	x	x
<b>High fidelity PCR</b>	x	x	✓	✓
<b>Blunt- end cloning</b>	x	x	✓	✓
<b>Site directed mutagenesis</b>	x	x	✓	✓
<b>Equipment &amp; Reagents to be supplied by user</b>				
<ul style="list-style-type: none"> <li>• Pipets and pipet tips</li> <li>• Microcentrifuge tube</li> <li>• Thermal cycler</li> <li>• Mineral oil (for thermal cyclers without a heated lid)</li> <li>• Primers</li> </ul>				

- \* Since, it is not hot-start, we recommended to add enzyme last during PCR.
- \*\* PCR Product can be directly cloned in to Blunt Vectors.
- \*\*\* compare to Taq DNA Polymerase
- \*\*\*\* Template-independent 'A' can be generated at the 3' end of PCR product.

## Prepare PCR Reactions

It is recommended to prepare PCR reactions as indicated in Table2.

**Table 2.** Prepare PCR Reactions, 5t-pfu PCR Set

Component	Volume
Template	Variable
Forward primer (10 µM)	1 µl
Reverse primer (10 µM)	1 µl
5t-Pfu Polymerase, Recombinant, (2.5U/µl)	1 µl
PCR Buffer, 10X, with MgSO <sub>4</sub> , Optimized for 5t-Pfu Polymerase	4 µl
HiPure-dNTPs mix, 2.5 mM	5-8 µl
Water for Molecular Biology, Sterile, Filtered	Up to 50 µl

## Tips for Optimizing PCR Reaction

- A final concentration of 2mM MgSO<sub>4</sub> is sufficient for most targets amplification. For some targets, more Mg<sup>2+</sup> may be required; use the 50 mM MgSO<sub>4</sub> stock to test from 2 mM to 4 mM (final concentration) in 0.25 mM increments.