










# PCR SuperMix

	<b>Package Contents</b>	<b>Catalog Number</b> 10572-014 10572-063	<b>Size</b> 100 rxns 5,000 rxns	 Kit Contents
	<b>Storage Conditions</b>	<ul style="list-style-type: none"> <li>Store all contents at -20°C.</li> </ul>		
	<b>Required Materials</b>	<ul style="list-style-type: none"> <li>Template: cDNA, gDNA, λDNA</li> <li>Forward and reverse gene-specific primers</li> <li>Autoclaved, distilled water</li> <li>E-Gel® Gels, 1.2% (Cat. no. G5018-01)</li> <li>TrackIt™ 1 Kb Plus DNA Ladder (Cat. no. 10488-085)</li> <li>0.2 or 0.5-mL nuclease-free microcentrifuge tubes</li> </ul>		
	<b>Timing</b>	Varies depending on amplicon length		
	<b>Selection Guide</b>	<a href="#">PCR Enzymes and Master Mixes</a> Go online to view related products.		
	<b>Product Description</b>	<ul style="list-style-type: none"> <li>PCR SuperMix contains Mg<sup>++</sup>, dNTPs, and recombinant <i>Taq</i> DNA polymerase.</li> <li>PCR SuperMix is supplied at 1.1X concentration to allow approximately 10% of the final reaction volume to be used for primer and template solutions.</li> <li>A standard 50-μL reaction uses 45 μL of PCR SuperMix and 5 μL of primer and template solution.</li> </ul>		
	<b>Important Guidelines</b>	<ul style="list-style-type: none"> <li>Select the correct polymerase, PCR instrument, and cycling conditions for your application.</li> <li>Take precautions to avoid cross-contamination by using aerosol-resistant barrier tips and analyzing PCR products in a separate area from PCR assembly.</li> <li>If PCR efficiency is sub-optimal, repeat the reaction with different primer concentrations from 0.1–0.5 μM (final).</li> <li>If the final Mg<sup>++</sup> concentration is adjusted to 1.5 mM, the volume of primer and template solution that can be added to 45 μL of PCR SuperMix can exceed 50 μL.</li> </ul>		
	<b>Online Resources</b>	Visit our <a href="#">product page</a> for additional information and protocols. For support, visit <a href="http://www.lifetechnologies.com/support">www.lifetechnologies.com/support</a> .		



## Enzyme Characteristics


<b>Hot-start:</b>	None
<b>Length:</b>	Up to 5 kb
<b>Fidelity vs. <i>Taq</i>:</b>	1X
<b>Format:</b>	Master mix

## PCR Reaction Setup

Use the measurements below to prepare your PCR experiment, or enter your own parameters in the column provided.

Component	25-μL rxn	50-μL rxn	Custom	Final Conc.
Autoclaved, distilled water	to 25 μL	to 50 μL	to μL	–
PCR SuperMix	22.5 μL	45 μL	μL	1X
10 μM forward primer	0.5 μL	1 μL	μL	0.2 μM
10 μM reverse primer	0.5 μL	1 μL	μL	0.2 μM
Template DNA	varies	varies		< 500 ng

## PCR Protocol

 See page 2 to view a procedure for preparing and running your PCR experiment.

## Optimization Strategies





 Refer to the pop-up for guidelines to optimize your PCR reactions.

## Limited Warranty, Disclaimer, and Licensing Information

## PCR SuperMix Protocol

The example PCR procedure below shows appropriate volumes for a single **50- $\mu$ L** reaction.

For multiple reactions, prepare a master mix of components common to all reactions to minimize pipetting error, and then dispense appropriate volumes into each 0.2–0.5 mL PCR reaction tube prior to adding template DNA and primers.

Timeline		Steps	Procedure Details																		
1		<b>Thaw reagents</b>	<p>Thaw, mix, and briefly centrifuge each component before use.</p> <p>Set up the reaction tubes/plates on ice.</p> <p>Add the following components to each PCR reaction tube.</p> <p>Adjust the reaction volumes as needed for your application.</p>																		
2		<b>Add template DNA and primers to mix</b>	<table border="1"> <thead> <tr> <th>Component</th> <th>50-<math>\mu</math>L rxn</th> <th>Final Concentration</th> </tr> </thead> <tbody> <tr> <td>Autoclaved, distilled water</td> <td>to 50 <math>\mu</math>L</td> <td></td> </tr> <tr> <td>PCR SuperMix</td> <td>45 <math>\mu</math>L</td> <td>1X</td> </tr> <tr> <td>10 uM forward primer</td> <td>1 <math>\mu</math>L*</td> <td>0.2 <math>\mu</math>M</td> </tr> <tr> <td>10 uM reverse primer</td> <td>1 <math>\mu</math>L*</td> <td>0.2 <math>\mu</math>M</td> </tr> <tr> <td>Template DNA</td> <td>varies*</td> <td>&lt; 500 ng</td> </tr> </tbody> </table> <p>* The total volume of primer and template solution should be 0.5–20 <math>\mu</math>L. If the final Mg concentration is adjusted to 1.5 mM, the volume of primer and template solution that can be added to 45 <math>\mu</math>L of PCR SuperMix can exceed 50 <math>\mu</math>L.</p>	Component	50- $\mu$ L rxn	Final Concentration	Autoclaved, distilled water	to 50 $\mu$ L		PCR SuperMix	45 $\mu$ L	1X	10 uM forward primer	1 $\mu$ L*	0.2 $\mu$ M	10 uM reverse primer	1 $\mu$ L*	0.2 $\mu$ M	Template DNA	varies*	< 500 ng
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3		<b>Incubate reactions in a thermal cycler</b>	<p>Cap each tube, mix, and then briefly centrifuge the contents.</p> <table border="1"> <thead> <tr> <th>Step</th> <th>Temperature (<math>^{\circ}</math>C)</th> <th>Time</th> </tr> </thead> <tbody> <tr> <td>Initial Denaturation</td> <td>94</td> <td>2 minutes</td> </tr> <tr> <td rowspan="3">25–35 PCR Cycles</td> <td>Denature</td> <td>94</td> </tr> <tr> <td>Anneal</td> <td>~55 (depending on primer <math>T_m</math>)</td> <td>30 seconds</td> </tr> <tr> <td>Extend</td> <td>72</td> <td>1 minute/kb</td> </tr> <tr> <td>Hold</td> <td>4</td> <td>indefinitely</td> </tr> </tbody> </table>	Step	Temperature ( $^{\circ}$ C)	Time	Initial Denaturation	94	2 minutes	25–35 PCR Cycles	Denature	94	Anneal	~55 (depending on primer $T_m$ )	30 seconds	Extend	72	1 minute/kb	Hold	4	indefinitely
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4		<b>Analyze with gel electrophoresis</b>	<p>Analyze 10 <math>\mu</math>L using agarose gel electrophoresis.</p> <p>Use your PCR reaction immediately for down-stream applications, or store it at <math>-20^{\circ}</math>C.</p>																		