





Platinum® *Taq* DNA Polymerase High Fidelity


	Package Contents	Catalog Number	Size	 Kit Contents
		11304-011	100 rxns	
		11304-029	500 rxns	
		11304-102	5,000 rxns	


	Storage Conditions	<ul style="list-style-type: none"> Store all contents at -20°C.
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	Required Materials	<ul style="list-style-type: none"> Template: cDNA, gDNA, λDNA Forward and reverse gene-specific primers 10 mM dNTP mix (Cat. no. 18427-088) Autoclaved, distilled water E-Gel® General Purpose Gels, 1.2% (Cat. no. G5018-01) TrackIt™ 1 Kb Plus DNA Ladder (Cat. no. 10488-085) 0.2 or 0.5-mL nuclease-free microcentrifuge tubes
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	Timing	Varies depending on amplicon length
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	Selection Guide	<p>PCR Enzymes and Master Mixes</p> <p>Go online to view related products.</p>
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	Product Description	<ul style="list-style-type: none"> Platinum® <i>Taq</i> DNA Polymerase High Fidelity contains recombinant <i>Taq</i> DNA polymerase, <i>Pyrococcus species</i> GB-D polymerase, and Platinum® <i>Taq</i> Antibody. This enzyme allows amplification of simple and complex DNA templates over a large range of target sizes and provides 6X higher fidelity over <i>Taq</i>. Activity is restored after the denaturation step in PCR cycling at 94°C, providing an automatic “hot start” and offering increased sensitivity, specificity, and yield, while allowing assembly of reactions at room temperature.
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	Important Guidelines	<ul style="list-style-type: none"> Select the correct polymerase, PCR instrument, and cycling conditions for your application. Take precautions to avoid cross-contamination by using aerosol-resistant barrier tips and analyzing PCR products in a separate area from PCR assembly. For gDNA or cDNA, use a primer concentration of 0.2 μM. For Plasmid or λDNA, increase to 0.4 μM. Do not perform the initial denaturation for more than 30 seconds if the target is greater than 12 kb.
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	Online Resources	Visit our product page for additional information and protocols. For support, visit www.lifetechnologies.com/support .
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Enzyme Characteristics


Hot-start:	Antibody
Length:	Up to 20 kb
Fidelity vs. <i>Taq</i>:	6X
Format:	Separate components

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	–
10X High Fidelity PCR Buffer	2.5 μL	5 μL	μL	1X
50 mM MgSO ₄	1 μL	2 μL	μL	2.0 mM
10 mM dNTP Mix	0.5 μL	1 μL	μL	0.2 mM each
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
Platinum® <i>Taq</i> DNA Polymerase High Fidelity (5 U/μL)	0.1 μL	0.2 μL	μL	1 U/rxn

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**

Platinum® Taq DNA Polymerase High Fidelity Protocol

The example PCR procedure below shows appropriate volumes for a single 50- μ 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		Thaw reagents	Thaw, mix, and briefly centrifuge each component before use																																														
2		Prepare PCR master mix	<p>Add the following components to each PCR reaction tube.</p> <p>Note: Consider the volumes for all components listed in steps 2 and 3 to determine the correct amount of water required to reach your final reaction volume.</p> <table border="1"> <thead> <tr> <th>Component</th> <th>50-μL rxn</th> <th>Final Concentration</th> </tr> </thead> <tbody> <tr> <td>Autoclaved, distilled water</td> <td>to 50 μL</td> <td></td> </tr> <tr> <td>10X High Fidelity PCR Buffer</td> <td>5 μL</td> <td>1X</td> </tr> <tr> <td>10 mM dNTP mix</td> <td>1 μL</td> <td>0.2 mM each</td> </tr> <tr> <td>50 mM MgSO₄</td> <td>2 μL</td> <td>2 mM</td> </tr> <tr> <td>Platinum® Taq DNA High Fidelity Polymerase (5 U/μL)</td> <td>0.2 μL</td> <td>1 U*</td> </tr> </tbody> </table> <p>* 1 U is sufficient for amplifying most targets. In some cases, more enzyme may be required (up to 2.5 units). Use 1 U of enzyme for targets > 12 kb.</p> <p>Mix and briefly centrifuge the components.</p> <p>Add your template DNA and primers to each tube for a final reaction volume of 50 μL.</p> <table border="1"> <thead> <tr> <th>Component</th> <th>50-μL rxn</th> <th>Final Concentration</th> </tr> </thead> <tbody> <tr> <td>10 μM forward primer</td> <td>1–2 μL</td> <td>0.2 μM (Genomic and cDNA)– 0.4 μM (Plasmid and λDNA)</td> </tr> <tr> <td>10 μM reverse primer</td> <td>1–2 μL</td> <td>0.2 μM (Genomic and cDNA)– 0.4 μM (Plasmid and λDNA)</td> </tr> <tr> <td>Template DNA</td> <td>varies</td> <td>< 500 ng</td> </tr> </tbody> </table> <p>Cap each tube, mix, and then briefly centrifuge the contents.</p> <table border="1"> <thead> <tr> <th>Step</th> <th>Temperature (°C)</th> <th>Time</th> </tr> </thead> <tbody> <tr> <td>Initial Denaturation</td> <td>94</td> <td>30 seconds–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 T_m)</td> </tr> <tr> <td>Extend</td> <td>68</td> </tr> <tr> <td>Hold</td> <td>4</td> <td>indefinitely</td> </tr> </tbody> </table> <p>* For targets > 12 kb, do not exceed 30 seconds for initial denaturation.</p>	Component	50- μ L rxn	Final Concentration	Autoclaved, distilled water	to 50 μ L		10X High Fidelity PCR Buffer	5 μ L	1X	10 mM dNTP mix	1 μ L	0.2 mM each	50 mM MgSO ₄	2 μ L	2 mM	Platinum® Taq DNA High Fidelity Polymerase (5 U/ μ L)	0.2 μ L	1 U*	Component	50- μ L rxn	Final Concentration	10 μ M forward primer	1–2 μ L	0.2 μ M (Genomic and cDNA)– 0.4 μ M (Plasmid and λ DNA)	10 μ M reverse primer	1–2 μ L	0.2 μ M (Genomic and cDNA)– 0.4 μ M (Plasmid and λ DNA)	Template DNA	varies	< 500 ng	Step	Temperature (°C)	Time	Initial Denaturation	94	30 seconds–2 minutes*	25–35 PCR Cycles	Denature	94	Anneal	~55 (depending on primer T _m)	Extend	68	Hold	4	indefinitely
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4		Incubate reactions in a thermal cycler																																															
5		Analyze with gel electrophoresis	<p>Analyze 10 μL using agarose gel electrophoresis.</p> <p>Use your PCR reaction immediately for down-stream applications, or store it at -20°C.</p>																																														