

	Catalog number	Size	
Package contents	12369010	100 reactions	Kit contents
	12369050	500 reactions	
	12369250	5 × 500 reactions	

Storage conditions	<ul style="list-style-type: none"> Store all contents at -20°C.
---------------------------	---

Required materials	Click here for required materials
---------------------------	-----------------------------------

- Platinum™ SuperFi™ II DNA Polymerase is a proofreading DNA polymerase that combines high fidelity with Platinum™ hot-start technology and universal primer annealing. It is ideal for cloning, mutagenesis, and other applications.
- Platinum™ SuperFi™ II Green PCR Master Mix is a ready-to-use mixture of DNA polymerase, salts, magnesium, and dNTPs for efficient PCR amplification, which retains all the features of the Platinum™ SuperFi™ II DNA Polymerase. In addition, it is supplemented with two tracking dyes and a density reagent for direct loading of PCR products on gels.
- The annealing temperature with Platinum™ SuperFi™ II DNA Polymerase is 60°C . Proprietary additives in the reaction buffer stabilize primer-template duplexes during the annealing step, and contribute to increased specificity without the need to optimize annealing temperature for each primer pair.
- Due to proprietary additives in the reaction buffer, Platinum™ SuperFi™ II DNA Polymerase shows efficient amplification of both AT and GC rich targets. Additional DNA melting agents are not required for GC-rich PCR (up to 75% GC).
- Platinum™ hot-start technology inhibits DNA polymerase activity at ambient temperatures, allowing room temperature reaction setup and storage of pre-assembled PCR reactions. Enzyme activity is restored after the initial denaturation step.
- Platinum™ SuperFi™ II DNA Polymerase has 5' to 3' polymerase and 3' to 5' exonuclease activities, but lacks 5' to 3' exonuclease activity. It produces blunt end DNA products.

Product description	
----------------------------	--

Selection guide	PCR Enzymes and Master Mixes Go online to view related products.
------------------------	---

Online resources	Visit our product page for additional information and protocols. For support, visit thermofisher.com/support .
-------------------------	---

Enzyme characteristics

Hot-start:	Antibody
Length:	Up to 20 kb
Fidelity vs. <i>Taq</i>:	>300X
Timing:	Varies depending on amplicon length
Format:	Master Mix

PCR setup

Component	Final concentration	20- μL rxn	50- μL rxn
2X Platinum™ SuperFi™ II Green PCR Master Mix ^[1]	1X	10 μL	25 μL
Forward primer	0.5 μM ^[2]	x μL	x μL
Reverse primer	0.5 μM ^[2]	x μL	x μL
Template DNA	0.1–10 ng plasmid (5–100 ng genomic DNA)	x μL	x μL
Water, nuclease-free	—	to 20 μL	to 50 μL

^[1] Provides 1.75 mM MgCl_2 in 1X concentration.

^[2] Reduce the primer concentration to 0.2 μM final for amplification of >5 kb targets from genomic DNA and for multiplex reactions.

PCR protocol

See page 2 to prepare and run your PCR experiment.

Important guidelines

Click here for important PCR guidelines.

Optimization strategies and troubleshooting

Click here for guidelines to optimize your PCR experiment.




Click here for guidelines to troubleshoot your PCR experiment.


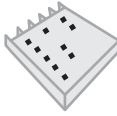
Purchaser notification

Click here for Limited Warranty, Disclaimer, and Licensing information.

Prepare and run PCR

The example PCR procedure below shows appropriate volumes for a single **20- μ L** or **50- μ L** reaction. For multiple reactions, prepare a master mix of components common to all reactions to minimize pipetting error, then dispense appropriate volumes into each 0.2-mL or 0.5-mL PCR tube before adding template DNA and primers.

Steps	Action	Procedure details																										
1 	Thaw reagents	Thaw, mix, and briefly centrifuge each component before use.																										
2 	Prepare reaction mix with template DNA and primers	<p>Add the following components to each PCR tube.</p> <p>Note: Consider the volumes for all components in reaction mix to determine the correct amount of water required to reach your final reaction volume.</p> <table border="1"> <thead> <tr> <th>Component</th> <th>Final concentration</th> <th>20-μL rxn</th> <th>50-μL rxn</th> </tr> </thead> <tbody> <tr> <td>2X Platinum™ SuperFi™ II Green PCR Master Mix ^[1]</td> <td>1X</td> <td>10 μL</td> <td>25 μL</td> </tr> <tr> <td>Forward primer</td> <td>0.5 μM ^[2]</td> <td>x μL</td> <td>x μL</td> </tr> <tr> <td>Reverse primer</td> <td>0.5 μM ^[2]</td> <td>x μL</td> <td>x μL</td> </tr> <tr> <td>Template DNA</td> <td>0.1–10 ng plasmid (5–100 ng genomic DNA)</td> <td>x μL</td> <td>x μL</td> </tr> <tr> <td>Water, nuclease-free</td> <td>—</td> <td>to 20 μL</td> <td>to 50 μL</td> </tr> </tbody> </table> <p>^[1] Provides 1.75 mM MgCl₂ in 1X concentration. ^[2] Reduce the primer concentration to 0.2 μM final for amplification of >5 kb targets from genomic DNA and for multiplex reactions.</p> <p>Cap each tube, mix, and then briefly centrifuge the contents.</p>	Component	Final concentration	20- μ L rxn	50- μ L rxn	2X Platinum™ SuperFi™ II Green PCR Master Mix ^[1]	1X	10 μ L	25 μ L	Forward primer	0.5 μ M ^[2]	x μ L	x μ L	Reverse primer	0.5 μ M ^[2]	x μ L	x μ L	Template DNA	0.1–10 ng plasmid (5–100 ng genomic DNA)	x μ L	x μ L	Water, nuclease-free	—	to 20 μ L	to 50 μ L		
Component	Final concentration	20- μ L rxn	50- μ L rxn																									
2X Platinum™ SuperFi™ II Green PCR Master Mix ^[1]	1X	10 μ L	25 μ L																									
Forward primer	0.5 μ M ^[2]	x μ L	x μ L																									
Reverse primer	0.5 μ M ^[2]	x μ L	x μ L																									
Template DNA	0.1–10 ng plasmid (5–100 ng genomic DNA)	x μ L	x μ L																									
Water, nuclease-free	—	to 20 μ L	to 50 μ L																									
3 	Incubate reactions in a thermal cycler (3-step protocol)	<p>3-step protocol</p> <table border="1"> <thead> <tr> <th>Cycle step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Initial denaturation</td> <td>98°C</td> <td>30 seconds</td> <td>1</td> </tr> <tr> <td>Denaturation</td> <td>98°C</td> <td>5–10 seconds</td> <td rowspan="3">25–35</td> </tr> <tr> <td>Annealing</td> <td>60°C</td> <td>10 seconds</td> </tr> <tr> <td>Extension</td> <td>72°C</td> <td>15–30 seconds per 1 kb</td> </tr> <tr> <td>Final extension</td> <td>72°C</td> <td>5 minutes</td> <td>1</td> </tr> <tr> <td></td> <td>4°C</td> <td>Hold</td> <td>—</td> </tr> </tbody> </table> <p>Note: Refer to “Optimization strategies”, page 1, for guidelines to optimize cycling conditions.</p>	Cycle step	Temperature	Time	Cycles	Initial denaturation	98°C	30 seconds	1	Denaturation	98°C	5–10 seconds	25–35	Annealing	60°C	10 seconds	Extension	72°C	15–30 seconds per 1 kb	Final extension	72°C	5 minutes	1		4°C	Hold	—
Cycle step	Temperature	Time	Cycles																									
Initial denaturation	98°C	30 seconds	1																									
Denaturation	98°C	5–10 seconds	25–35																									
Annealing	60°C	10 seconds																										
Extension	72°C	15–30 seconds per 1 kb																										
Final extension	72°C	5 minutes	1																									
	4°C	Hold	—																									

Steps	Action	Procedure details																			
<p>3</p> 	<p>Incubate reactions in a thermal cycler (2-step protocol)</p>	<p>2-step protocol (for primers >30 nt long)^[1]</p> <table border="1" data-bbox="707 214 1669 469"> <thead> <tr> <th>Cycle step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Initial denaturation</td> <td>98°C</td> <td>30 seconds</td> <td>1</td> </tr> <tr> <td>Denaturation</td> <td>98°C</td> <td>5–10 seconds</td> <td rowspan="2">25–35</td> </tr> <tr> <td>Annealing/Extension</td> <td>72°C</td> <td>15–30 seconds per 1 kb</td> </tr> <tr> <td>Final extension</td> <td>72°C 4°C</td> <td>5 minutes hold</td> <td>1 —</td> </tr> </tbody> </table> <p>^[1] Without non-complementary parts (e.g. restriction tags).</p> <p>Note: Refer to “Optimization strategies”, page 1, for guidelines to optimize cycling conditions.</p>	Cycle step	Temperature	Time	Cycles	Initial denaturation	98°C	30 seconds	1	Denaturation	98°C	5–10 seconds	25–35	Annealing/Extension	72°C	15–30 seconds per 1 kb	Final extension	72°C 4°C	5 minutes hold	1 —
Cycle step	Temperature	Time	Cycles																		
Initial denaturation	98°C	30 seconds	1																		
Denaturation	98°C	5–10 seconds	25–35																		
Annealing/Extension	72°C	15–30 seconds per 1 kb																			
Final extension	72°C 4°C	5 minutes hold	1 —																		
<p>4</p> 	<p>Add gel loading buffer and analyze with gel electrophoresis</p>	<p>Analyze the sample using agarose gel electrophoresis.</p> <p>Note: For optimal separation using E-Gel™ agarose gels, dilute the sample 2- to 20-fold.</p> <p>Note: PCR mixes prepared using the Platinum™ SuperFi™ II Green PCR Master Mix are ready for direct loading on the gels; addition of loading buffer is not needed.</p> <p>Use your PCR product immediately in down-stream applications, or store it at –20°C.</p>																			