# Invitrogen™ Platinum™ SuperFi™ Green PCR Master Mix

invitrogen

USER	GUIDE		Pub. No. MAN0014	885 <b>Rev</b> .	. В			
		Catalog number	Size					
	Package contents	12359-010	100 rxns	<b>f</b> Kit content	Lo.			
		12359-050	500 rxns	Kit content	21115			
		12359-250	$5 \times 500 \text{ rxns}$					
	Storage conditions	<ul> <li>Store all content</li> </ul>	Store all contents at –20°C.					
		ge DNA, cDNA						





 Forward and reverse gene-specific primers Invitrogen<sup>™</sup> E-Gel<sup>™</sup> General Purpose Gels, 1.2% (Cat. No. G501801)

Invitrogen<sup>™</sup> TrackIt<sup>™</sup> 1 kb Plus DNA Ladder (Cat. No. 10488085)

• 0.2 or 0.5-mL nuclease-free microcentrifuge tubes



#### **Timing**

Varies depending on amplicon length.



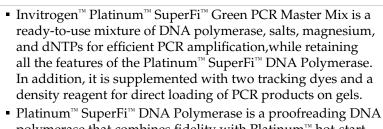
#### Selection quide

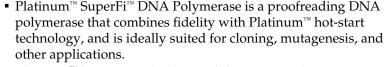
Product

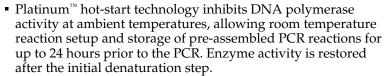
description

PCR Enzymes and Master Mixes

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- Platinum<sup>™</sup> SuperFi<sup>™</sup> 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.
- Platinum<sup>™</sup> SuperFi<sup>™</sup> PCR Master Mix is supplied with a separate vial of SuperFi<sup>™</sup> GC Enhancer designed for GC-rich templates (>65% GC).

## Important guidelines



Click here for important PCR guidelines.



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. *Taq*: >100X

Format: Master Mix

#### PCR setup

Component	25-μL rxn	50-μL rxn	i0-μL rxn Custom		Final conc.	
Water, nuclease-free	to 25 μL	to 50 µL	to	μL	_	
2X Platinum <sup>™</sup> SuperFi <sup>™</sup> Green PCR Master Mix <sup>1</sup>	12.5 μL	25 μL		μL	1X	
10 μM forward primer	1.25 μL	2.5 µL		μL	0.5 μΜ	
10 μM reverse primer	1.25 µL	2.5 µL		μL	0.5 μΜ	
Template DNA <sup>2</sup>	varies	varies			varies	
5X SuperFi <sup>™</sup> GC Enhancer (optional) <sup>3</sup>	5 μL	10 μL		μL	1X	

<sup>&</sup>lt;sup>1</sup> Provides MgCl, at a final concentration of 1.5 mM in the reaction.

### PCR protocol

See page 2 and page 3 to prepare and run your PCR experiment.

#### Optimization strategies and troubleshooting

Click here for guidelines to optimize your PCR experiment.

1 Click here for guidelines to troubleshoot your PCR experiment.

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<sup>&</sup>lt;sup>2</sup> 5–50 ng gDNA or 1 pg–10 ng plasmid DNA (see "**Optimization strategies**", below, for more information).

<sup>&</sup>lt;sup>3</sup> Recommended for targets with >65% GC sequences.

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, then dispense appropriate volumes into each 0.2–0.5 mL PCR tube before adding template DNA and primers.

Steps Action		Action	Procedure details						
1		Thaw reagents	Thaw, mix, and briefly centrifuge each component before use. Avoid generating bubbles when mixing the Master Mix.						
		Prepare PCR master mix	Add the following components to each PCR tube.  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.						
			Component	50-μL rxn	Final conc.				
2			Water, nuclease-free	to 50 µL					
			2X Platinum™ SuperFi™ Green PCR Master Mix	25 µL	1X				
			5X SuperFi™ GC Enhancer (optional)¹	10 μL	1X				
			<sup>1</sup> Recommended for targets with >65% GC sequences.  Mix and then briefly centrifuge the components.						
		Add template DNA and primers	Add your template DNA and primers to each tube for a final reaction volume of 50 µL.						
			Component 50-µL	rxn Final co	onc.				
	1 9 1		10 μM forward primer 2.5 μl	0.5 μΝ	Л				
3	8		10 μM reverse primer 2.5 μl	. 0.5 μΝ	М				
			Template DNA <sup>1</sup> varie	s varie	S				
			<ul> <li>Optimal amount of low complexity DNA (plasmid, phage, BAC DNA) is 1 pg–10 ng per 50 μL reaction, but it can be varied from 0.1 pg to 50 ng per 50 μL reaction. Optimal amount of genomic DNA is 5–50 ng per 50 μL reaction, but it can be varied from 0.1 ng to 250 ng per 50 μL reaction.</li> <li>Cap each tube, mix, and then briefly centrifuge the contents.</li> </ul>						

	Steps	Action	Procedure details							
4		Incubate reactions in a thermal cycler	Initial d  25–35 PCR cycles Final ex	ANT! Always t	Temp.  98°C  98°C  —  72°C  72°C  4°C  use T <sub>m</sub> calculator annealing	g temperature.	Temp. 98°C 98°C varies 72°C 72°C 4°C at www.therm	tocol (<10kb) Time 30 sec 5–10 sec 10 sec 15–30 sec/kb 5 min hold ofisher.com/tmca	Temp. 95°C 95°C varies 68°C 68°C 4°C	m ·
5		Analyze with gel electrophoresis	Analyze the sample using agarose gel electrophoresis.  Note: PCR mixes prepared using the SuperFi™ Green Buffer are ready for direct loading on the gels; addition of loading buffer is not needed.  Use your PCR product immediately in down-stream applications, or store it at −20°C.							

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