### Invitrogen™ Platinum™ Green Hot Start PCR 2X Master Mix



Pub. no. MAN0014005 Rev. A.0

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### Package contents

 Catalog number
 Size

 13001-012
 50 rxns

 13001-013
 200 rxns

 13001-014
 1000 rxns





## Storage conditions

- Store all contents at –20°C.
- Template: cDNA, gDNA, λDNA
- Forward and reverse gene-specific primers



- Invitrogen™ E-Gel™ General Purpose Gels, 1.2% (Cat. no. G5018-01)
- Invitrogen<sup>™</sup> TrackIt<sup>™</sup> 1 kb Plus DNA Ladder (Cat. no. 10488-085)
- 0.2 or 0.5-mL nuclease-free microcentrifuge tubes



#### **Timing**

Varies depending on amplicon length



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Product

description

PCR Enzymes and Master Mixes

Go online to view related products.

- Platinum<sup>™</sup> Green Hot Start PCR 2X Master Mix contains Platinum<sup>™</sup> Taq DNA Polymerase in an optimized PCR buffer with Mg<sup>2+</sup> and dNTPs. The master mix is supplemented with tracking dyes for direct loading of PCR products on gels.
- The master mix retains all features of the Platinum<sup> $\mathsf{T}$ </sup>  $\mathsf{Taq}$  DNA Polymerase.
- Specific binding of the anti-*Taq* antibody inhibits polymerase activity at ambient temperatures. Activity is restored after the initial denaturation step in PCR cycling at 94°C, providing an automatic "hot start" and offering increased sensitivity, specificity, and yield, while allowing reaction assembly at room temperature.
- Platinum<sup>™</sup> GC Enhancer improves amplification of GC-rich targets.
- The tracking dyes (a blue dye and a yellow dye) in the master mix do not interfere with PCR performance and are compatible with downstream applications such as fluorescent automatic DNA sequencing, ligation, and restriction digestion.



### Important guidelines

Click here for important PCR guidelines.



### Online resources

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

#### For Research Use Only. Not for use in diagnostic procedures.

#### **Enzyme characteristics**

Hot-start: Antibody Length: Up to 5 kb

Fidelity vs. *Taq*: 1X

**Format:** Master mix

#### **PCR** 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. in 50-µL rxn	
Water, Nuclease-free	to 25 μL	to 50 µL	to	μL	<del></del>	
Platinum <sup>™</sup> Green Hot Start PCR 2X Master Mix	12.5 μL	25 μL		μL	1X*	
10 μM forward primer	0.5 µL	1 μL		μL	0.2 μΜ	
10 µM reverse primer	0.5 µL	1 μL	μL		0.2 μΜ	
Template DNA	varies	varies			<500 ng/rxn	
Platinum™ GC Enhancer (optional)**	5 μL	10 μL		μL	20%	

<sup>\* 1</sup>X master mix contains a final concentration of 1.5 mM MgCl<sub>2</sub> and 0.2 mM of each dNTP.

#### PCR protocol

**1** See page 2 for instructions to prepare and run your PCR experiment.

#### **Optimization strategies**

f Click here for guidelines to optimize your PCR experiment.

#### **Purchaser notification**

1 Click here for Limited warranty, Disclaimer, and Licensing information.



<sup>\*\*</sup> Recommended only for targets with >65% GC sequences.

The example PCR procedure below shows appropriate volumes for a single  $50-\mu L$  reaction using GC-rich template DNA. For multiple reactions, prepare a master mix of common reaction components, then dispense appropriate volumes into each 0.2–0.5 mL PCR tube prior to adding template DNA and primers.

	Steps	Action	Procedure details							
1		Thaw reagents	Thaw, mix, and briefly centrifuge each component before use. Avoid generating bubbles when mixing the Master Mix.							
2		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.								
	Prepare PCR master mix	Component			50-μL rxn	Final conc.				
		Water, Nucleas	Water, Nuclease-free			_				
		Platinum <sup>™</sup> Green Hot Start PCR 2X Master Mix			25 µL	1X				
		Platinum <sup>™</sup> GC Enhancer (optional)*			10 μL	20%				
		*For targets with >65% GC sequences.								
		Mix and then briefly centrifuge the components.								
3	300	Add template DNA and primers	Add your template DNA and  Component  10 µM forward primer  10 µM reverse primer  Template DNA  Cap each tube, mix, and ther		50-µL rxn 1 µL 1 µL varies briefly centrifuge t	Final conc.  0.2 µM  0.2 µM  <500 ng/rxn  ne contents.		L.		
			Step		Temperature		Time			
4	Incubate reactions in a thermal cycler	Initial denaturation		94°C		2 minutes				
		25–35 PCR cycles	Denature	94°C		30 seconds				
			Anneal	~55°C (depending on primer T <sub>m</sub> )		30 seconds				
			Extend	72°C		1 minute/kb				
			Hold		4°C		indefinitely			
5	W. C.	Analyze with gel electrophoresis	Analyze the sample using agarose gel electrophoresis.  Note: The samples 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.							