Invitrogen™ Platinum™ *Taq* Green Hot Start DNA Polymerase

Pub. no. MAN0014006 Rev. A.0

invitrogen

Print Options

Package contents

Catalog number	Size
11966-018	120 rxns
11966-026	300 rxns
11966-034	600 rxns
11966-083	5000 rxns

Kit contents

Storage conditions

- Store all contents at –20°C.

Required materials

- Template: cDNA, gDNA, λDNA
- Forward and reverse gene-specific primers
- Invitrogen™ 10 mM dNTP mix (Cat. no. 18427-088)
- Water, nuclease-free
- Invitrogen™ E-Gel™ General Purpose Gels, 1.2% (Cat. no. G5018-01)
- Invitrogen™ TrackIt™ 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

Selection guide

PCR Enzymes and Master Mixes

Go online to view related products.

Product description

- Platinum™ *Taq* DNA Polymerase is a recombinant *Taq* polymerase complexed with a proprietary antibody that blocks the 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.
- 10X Green PCR buffer is supplemented with two tracking dyes (a blue dye and a yellow dye) and a density reagent for direct loading of PCR products on gels. The dyes in the buffer do not interfere with PCR performance and are compatible with downstream applications such as fluorescent automatic DNA sequencing, ligation, and restriction digestion.
- This enzyme has a non-template-dependent, terminal transferase activity that adds a single deoxyadenosine (A) to the 3' ends of PCR products. Like standard *Taq* polymerase, it has both 5' to 3' polymerase and 5' to 3' exonuclease activity, but lacks 3' to 5' exonuclease activity.

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: Separate components

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	—
10X Green PCR Buffer, – Mg	2.5 µL	5 µL	µL	1X
50 mM $MgCl_2$	0.75 µL	1.5 µL	µL	1.5 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	µL	<500 ng/rxn
KB Extender (*optional*)*	varies	varies	µL	3–9%
Platinum™ *Taq* DNA Polymerase	0.1 µL	0.2 µL	µL	2 U/rxn

* Recommended for targets >5 kb or with >65% GC sequences.

PCR protocol

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

Optimization strategies

Click here for guidelines to optimize your PCR experiment.

Purchaser notification

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

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The example PCR procedure below shows appropriate volumes for a single **50-µL** 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 prior to adding template DNA and primers.

Steps 1 & Action: Thaw reagents

Thaw, mix, and briefly centrifuge each component before use.

Steps 2 & Action: 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.
Water, nuclease-free	to 50 µL	
10X Green PCR Buffer, minus Mg	5 µL	1X
50 mM $MgCl_2$	1.5 µL	1.5 mM
10 mM dNTP mix	1 µL	0.2 mM each
KB Extender (*optional*)*	1.5–4.5 µL	3–9%
Platinum™ *Taq* DNA Polymerase	0.2 µL	2 U/rxn

*For targets >5 kb or with >65% GC sequences.

Mix and then briefly centrifuge the components.

Steps 3 & Action: 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 conc.
10 µM forward primer	1 µL	0.2 µM
10 µM reverse primer	1 µL	0.2 µM
Template DNA	varies	<500 ng/rxn

Cap each tube, mix, and then briefly centrifuge the contents.

Steps 4 & Action: Incubate reactions in a thermal cycler

Step		Temperature	Time
Initial denaturation		94°C	2 minutes
25–35 PCR cycles	Denature	94°C	30 seconds
	Anneal	~55°C (depending on primer T_m)	30 seconds
	Extend	72°C	1 minute/kb
Hold		4°C	indefinitely

Steps 5 & Action: Analyze with gel electrophoresis

Analyze the sample using agarose gel electrophoresis.

Note: PCR mixes prepared using the 10X Green PCR 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.