Platinum™ PCR SuperMix High Fidelity

Catalog Numbers 12532-016 and 12532-024

Doc. Part No. 12532.pps **Pub. No.** MAN0001086 **Rev.** A.0

WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **thermofisher.com/support**.

Product description

The Invitrogen™ Platinum™ PCR SuperMix High Fidelity is used for high-fidelity PCR amplification of DNA templates. It is effective over a large range of target sizes (up to 15 kb of genomic DNA). The mix contains anti-*Taq* DNA polymerase antibody, Mg^{++}, deoxyribonucleotide triphosphates, recombinant *Taq* DNA polymerase, and *Pyrococcus* species *GB-D* thermostable polymerase.

Anti-*Taq* DNA polymerase antibody inhibits polymerase activity, providing an automatic "hot start" and permitting room-temperature setup. Antibody-mediated hot starts improve PCR specificity and yield. The polymerase activity is restored after a denaturation step in PCR cycling at 94°C. *Pyrococcus* species *GB-D* polymerase possesses 3′ to 5′ exonuclease proofreading activity. Mixing the proofreading enzyme with *Taq* DNA polymerase increases fidelity ~6 times over that of *Taq* DNA polymerase alone.

The Platinum™ PCR SuperMix High Fidelity is supplied at a 1.1X concentration to allow ~10% of the final reaction volume to be used for the addition of primer and template solutions.

Contents and storage

Contents	Cat. No.		Storage
	12532-016 (100 rxns)	12532-024 (5,000 rxns)	
Platinum™ PCR SuperMix High Fidelity[1]	4 × 1.125 mL	4 × 56.25 mL	–20°C in a non-frost-free freezer[2]

[1] 22 U/mL of complexed recombinant *Taq* DNA polymerase, *Pyrococcus* species *GB-D* thermostable polymerase, and Platinum™ *Taq* Antibody; 66 mM Tris-SO_4 (pH 8.9); 19.8 mM $(NH_4)_2SO_4$; 2.4 mM $MgSO_4$; 220 µM dNTPs; and stabilizers.

[2] After thawing, store the mix at 4°C for 3 months or –20°C for 1 year. If you store the mix at 4°C, you do not have to thaw the mix before assembling the reactions. There is no detectable decrease in enzyme activity or performance after storage at 4°C for 3 months, or after 15 freeze-thaw cycles.

Procedural guidelines

- Take appropriate precautions to avoid cross-contamination.
- The reactions can be assembled at room temperature or on ice. We have observed no significant difference in reaction efficiency between these setup conditions.
- For multiple reactions, prepare a master mix of Platinum™ PCR SuperMix High Fidelity and the component(s) common to all reactions.
- If the PCR efficiency is not optimal, repeat the reaction with different primer concentrations. Start at 100 nM, then increase in 100 nM increments up to 500 nM.
- For longer genomic DNA targets (>15 kb), add 1–1.5 units of Platinum™ *Taq* DNA Polymerase (Cat. No. 10966-018) to the reaction mix and increase the extension time as specified (1 minute per kb).
- At higher volumes of primer and template, the $MgSO_4$ concentration in the reaction drops to suboptimal levels and yield decreases. For combined primer-template volumes of >15 µL (in solution with 45 µL of Platinum™ PCR SuperMix High Fidelity), adjust the final $MgSO_4$ concentration in the reaction to 2.2 mM.

Perform the PCR

1. Add the following components in any order to each reaction tube:

Component	Amount for one 50-µL reaction
Platinum™ PCR SuperMix High Fidelity	45 µL
Primer solution	200 nM final concentration of each is recommended
Template DNA solution	1–200 ng of genomic DNA

Note: A standard 50-µL PCR reaction includes a combined primer and template volume of 5 µL. We have observed no decrease in product yield if the amount of primer and template solution is between 1 µL and 15 µL.

2. Cap the tubes, tap gently to mix, then centrifuge briefly to collect the contents.
3. Load the tubes in a thermal cycler and incubate at 94°C for 30 seconds to 2 minutes to completely denature the template and activate the enzyme.
4. Perform 25–35 cycles of PCR amplification as follows:

Step	Temperature	Time
Denature	94°C	15–30 seconds
Anneal	55°C	15–30 seconds
Extend	68°C	1 minute per kb

5. Maintain the reactions at 4°C after cycling. Samples can be stored at –20°C until use.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.

The information in this guide is subject to change without notice.

DISCLAIMER

TO THE EXTENT ALLOWED BY LAW, LIFE TECHNOLOGIES AND/OR ITS AFFILIATE(S) WILL NOT BE LIABLE FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE, OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING YOUR USE OF IT.

Revision history: Pub. No. MAN0001086

Revision	Date	Description
A.0	5 May 2016	Format, style, and legal updates
—	11 May 2010	Baseline for this revision history

Corporate entity: Life Technologies Corporation | Carlsbad, CA 92008 USA | Toll Free in USA 1 800 955 6288