Platinum™ GenoType *Tsp* DNA Polymerase

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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™ GenoType *Tsp* DNA Polymerase is a recombinant DNA polymerase from a thermophilic species of bacteria. It is used to genotype dinucleotide repeat loci. The polymerase has been engineered to lack both 5′ and 3′ exonuclease activities and is severely restricted in its ability to add a nontemplated nucleotide to the end of the PCR product. It can be substituted directly for *Taq* DNA polymerase in amplification reactions as a simple solution to the heterogeneous extra nucleotide addition problem. It is recommended for amplification of fragments up to 500 bp in length.

The Platinum™ GenoType *Tsp* DNA Polymerase is supplied complexed with a proprietary antibody that inhibits polymerase activity at room temperature. The activity is regained after the denaturation step in PCR cycling at 94°C, providing an automatic "hot start". Hot starts are typically used in PCR to increase sensitivity, specificity, and yield while allowing assembly of reactions at ambient temperatures.

Contents and storage

Contents	Amount	Storage
Platinum™ GenoType *Tsp* DNA Polymerase (5 *Tsp* U/µL)	50 µL (250 *Tsp* U)	–20°C
10X PCR Buffer, Minus Mg[1]	1.25 mL	
50 mM Magnesium Chloride	1 mL	

[1] 200 mM Tris-HCl (pH 8.4), 500 mM KCl

Note: Unit (U) definition: One *Tsp* unit of Platinum™ GenoType *Tsp* DNA Polymerase has been functionally determined to be equivalent to one unit of *Taq* DNA polymerase for amplifying dinucleotide repeats using standard *Taq* reaction conditions.
One *Tsp* unit approximates 2.5 activity units. An activity unit incorporates 10 nmol of deoxyribonucleotide into acid–precipitable material in 30 minutes at 74°C under optimized reaction conditions.

Storage buffer

- 20 mM Tris-HCl (pH 8.0)
- 40 mM NaCl
- 2 mM Sodium Phosphate
- 0.1 mM EDTA
- 1 mM DTT
- Stabilizers
- 50% (v/v) glycerol

Perform the PCR

The following general procedure is suggested as a guideline and as a starting point when using Platinum™ GenoType *Tsp* DNA Polymerase in any PCR amplification.

1. Add the following components to the PCR reaction tube:

Component	Volume for one 15-µL reaction	Final concentration
10X PCR Buffer, Minus Mg	1.5 µL	1X
10 mM dNTP mixture	0.3 µL	0.2 mM each
50 mM $MgCl_2$	0.45 µL	1.5 mM
Primer mix (5 µM each)	1 µL	0.33 µM each
Template DNA	as required	50 ng
Platinum™ GenoType *Tsp* DNA Polymerase	0.12 µL	0.6 U
Autoclaved, distilled water	to 15 µL	—

Note: If needed, you can prepare a master mix for multiple reactions, to minimize reagent loss and to enable accurate pipetting.

2. Perform thermal cycling as follows:

No. of cycles	Step	Temperature	Time
1	(If needed) Predenaturation	94°C	1–2 minutes
10	Denature	94°C	30 seconds
	Anneal	55°C	30 seconds
	Extend	72°C	1 minute
20	Denature	89°C	30 seconds
	Anneal	55°C	30 seconds
	Extend	72°C	1 minute
1	(If needed) Final extension	72°C	10 minutes

3. Maintain the reactions at 4°C after cycling. The samples can be stored at –20°C until use.
4. Analyze the amplification products by electrophoresis. Use appropriate molecular weight standards to determine the size of the products.

Limited product warranty

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

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