

Platinum[®] GenoTYPE *Tsp* DNA Polymerase

Cat. No. 11448-024

Conc.: 5 *Tsp* U/ μ l

Size: 250 *Tsp* Units

**Store at -20°C in a
non-frost-free freezer.**

Description

Platinum[®] GenoTYPE *Tsp* DNA Polymerase is a recombinant DNA polymerase from a thermophilic species of bacteria. It is intended for use in genotyping of 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 heterogenous extra nucleotide addition problem. It is recommended for amplification of fragments up to 500 bp in length. The enzyme is supplied complexed with proprietary antibody that inhibits polymerase activity. Due to specific binding of the inhibitor, Platinum[®] GenoTYPE *Tsp* DNA Polymerase is provided in an inactive form. This reagent provides an automatic "hot start" for use in PCR (1,2). Hot starts are typically used in PCR to increase sensitivity, specificity and yield while allowing assembly of reactions at ambient temperatures. The extra time, effort, and contamination risks associated with manual hot start procedures are addressed with the use of Platinum[®] GenoTYPE *Tsp* DNA Polymerase. The activity of Platinum[®] GenoTYPE *Tsp* DNA Polymerase is blocked at ambient temperatures but is regained after the denaturation step in PCR cycling at 94°C.

Components

Platinum [®] GenoTYPE <i>Tsp</i> DNA Polymerase	11448-024
10X PCR Buffer, Minus Mg	Y02028
50 mM Magnesium Chloride	Y02016

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

Part no. 11448024.pps

MAN0000982

Rev. Date 07 Jun 2010

10X PCR Buffer

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

Unit Definition

One *Tsp* unit of Platinum® GenoTYPE *Tsp* DNA Polymerase has been functionally determined to be equivalent to one unit of *Taq* DNA Polymerase in amplification of 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 min at 74°C under optimized reaction conditions.

Product Qualification

The Certificate of Analysis (CofA) provides detailed quality control information for each product. The CofA is available on our website at www.invitrogen.com/cofa, and is searchable by product lot number, which is printed on each box.

Protocol

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:

<u>Components</u>	<u>Volume</u>	<u>Final Concentration</u>
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 <i>Tsp</i> DNA Polymerase	0.12 µl	0.6 units
Autoclaved, distilled water	to 15 µl	Not applicable

If desired, a master mix can be prepared for multiple reactions, to minimize reagent loss and to enable accurate pipetting.

2. Perform 30 cycles of PCR amplification as follows:
 - Pre-denaturation 94°C for 1-2 min (if desired)
 - Denature 94°C for 30 s
 - Anneal 55°C for 30 s
 - Extend 72°C for 1 min for 10 cycles
 - Denature 89°C for 30 s
 - Anneal 55°C for 30 s
 - Extend 72°C for 1 min for 20 cycles
 - Final extension 72°C for 10 min (if desired)
3. Maintain the reaction 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.

References

1. Chou, Q., *et al.* (1992) *Nucl. Acids Res.*, 20, 1717.
2. Sharkey, D.J., *et al.* (1994) *BioTechnology*, 12, 506.

Limited Use Label License No. 1: Thermostable Polymerases

Use of this product is covered by one or more of the following US patents and corresponding patent claims outside the U.S.: 5,789,224, 5,618,711, 6,127,155. The purchase of this product includes a limited, non-transferable immunity from suit under the foregoing patent claims for using only this amount of product for the purchaser's own internal research. No right under any other patent claim, no right to perform any patented method, and no right to perform commercial services of any kind, including without limitation reporting the results of purchaser's activities for a fee or other commercial consideration, is conveyed expressly, by implication, or by estoppel. This product is for research use only. Diagnostic uses under Roche patents require a separate license from Roche. Further information on purchasing licenses may be obtained by contacting the Director of Licensing, Applied Biosystems, 850 Lincoln Centre Drive, Foster City, California 94404, USA.

Limited Use Label License No. 5: Invitrogen Technology

The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes. The buyer may transfer information or materials made through the use of this product to a scientific collaborator, provided that such transfer is not for any Commercial Purpose, and that such collaborator agrees in writing (a) not to transfer such materials to any third party, and (b) to use such transferred materials and/or information solely for research and not for Commercial Purposes. Commercial Purposes means any activity by a party for consideration and may include, but is not limited to: (1) use of the product or its components in manufacturing; (2) use of the product or its components to provide a service, information, or data; (3) use of the product or its components for therapeutic, diagnostic or prophylactic purposes; or (4) resale of the product or its components, whether or not such product or its components are resold for use in research. For products that are subject to multiple limited use label licenses, the terms of the most restrictive limited use label license shall control. Life Technologies Corporation will not assert a claim against the buyer of infringement of patents owned or controlled by Life Technologies Corporation which cover this product based upon the manufacture, use or sale of a therapeutic, clinical diagnostic, vaccine or prophylactic product developed in research by the buyer in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product. If the purchaser is not willing to accept the limitations of this limited use statement, Life Technologies is willing to accept return of the product with a full refund. For information about purchasing a license to use this product or the technology embedded in it for any use other than for research use please contact Out Licensing, Life Technologies, 5791 Van Allen Way, Carlsbad, California 92008 ; Phone (760) 603-7200 or e-mail: outlicensing@lifetech.com.

Limited Use Label License No. 14: Direct Inhibition by Anti-Polymerase Antibodies

Licensed to Life Technologies Corporation, under U.S. Patent Nos. 5,338,671; 5,587,287, and foreign equivalents for use in research only.

Limited Use Label License No. 16: GenoTYPE™ Tsp

Purchase of this product does not convey to the purchaser a license to practice any claim of any third-party patent, including but not limited to U.S. Patents No. 5,766,847 and foreign equivalents. Users of this product should determine if any license is required under these or any other third-party patents.

©2010 Life Technologies Corporation. All rights reserved.

For research use only. Not intended for any animal or human therapeutic or diagnostic use.