UptiTherm DNA Polymerase - gel form

Product Description

Cat. No.	Product
	UptiTherm DNA Polymerase - Gel form
UPS54020.	250 units + Standard buffer (including 2 mM $MgCl_2$)
UPS54021	500 units + Standard buffer (including 2 mM $MgCl_2$)
UPS54031.	250 units + $MgCl_2$ free buffer + vial $MgCl_2$ 50 mM
UPS54032	500 units + $MgCl_2$ free buffer + vial $MgCl_2$ 50 mM
	UptiTherm DNA Polymerase - Gel Master Mix
UPS54071.	50 x 0,2 ml ($MgCl_2$ including vials) (50 µl final reaction volume, including 200µM dNTP, 1X buffer, 2mM $MgCl_2$ and 2U/well polymerase)
UPS54081	50 x 0,2 ml ($MgCl_2$ free vials) (50 µl final reaction volume, including 200µM dNTP, 1X buffer and polymerase; + $MgCl_2$ 50 mM vial)
S54090	12 x 8-tubes strips ($MgCl_2$ including vials)
SH7040	10 x 96-well plates ($MgCl_2$ including vials)
SH7041	20 x 96-well plates ($MgCl_2$ including vials) (including 10 mM dNTP, Reaction Buffer including 2mM $MgCl_2$ and 5U/well polymerase)

Also available with vials of Deoxynucleotide mix (Cat. Nos UPS54211, UPS54241)

Storage Conditions **Store at 4°C while in gel form (L)**
Store at -20°C once regenerated

Shipping can be performed at room temperature, as well as reaction setup, without the need of using ice.
Never freeze the gel at –20°C, as this will cause complete inactivation of the enzyme when in gel form.
Once the polymerase has been resuspended in the storage buffer, store at –20°C.

Scientific and Technical Information

Description

Highly thermostable DNA polymerase. It is a recombinant, modified form of the enzyme from the thermophilic bacterium *Thermus thermophilus* expressed in *E. coli* (see Note 1).

UptiTherm polymerase is suitable for applications which require a highly thermostable and processive enzyme capable of synthetising DNA strands at elevated temperatures in amplifications reactions or similar (e.g. primer extension), thus resolving the most complex secondary structures.

The enzyme is free of unspecific endo-or exonucleases activities, as well as nicking activities. It does not either exhibit signifiant reverse-transcriptase activity. Terminal transferase activity inherent to the enzyme renders A-tailed amplification products suitable to be further used in T/A cloning approaches.

The UptiTherm DNA Polymerase in gel form represents a step forward in respect to its liquid counterpart. It can be shipped at room temperature, and is stored at 4°C. This product has different presentations. On the one hand, there is polymerase in one vial, plus accompanying buffers (Standard or Mg-free). This format allows to have polymerase at the concentration desired, just resuspend in the appropriate amount of storage buffer provided with the polymerase, and proceed with your experiment (**after resuspension, store at –20°C**). The second

format is ready-to-use, aliquoted in 0,2 ml vials (including buffer, $MgCl_2$, dNTP). This way, only primers, your sample and water has to be added to the vial, minimising the handling steps and risks of contaminations.

For polymerase in vial format, and in order to perform hot-start reactions, it is not necessary to resuspend the vials' content, as reagents will only resuspend when 94°C are archieved. We recommend a preincubation step at 85°C for 2 min, followed by an initial denaturing step at 94°C for 2 min, in order to archieve the maximum yield in hot-start reactions.

Unit Definition

One unit is defined as the amount of enzyme which incorporates 10 nanomoles of dNTPs into acid-insoluble DNA within 30 minutes at 72 °C.

Reaction buffer

Recommended reaction buffer is : 75 mM Tris HCl (pH 9.0), **2 mM $MgCl_2$** (see Note 2), 50 mM KCl, 20mM $(NH_4)_2SO_4$. This reaction buffer (the so-called **Standard Buffer**, Ordering Information at the top) is supplied at 10X concentration together with the enzyme.
Reaction buffer can be supplied $MgCl_2$ free (see Ordering Information) : Mg^{2+} ion, being the enzyme cofactor, plays a key role on polymerase activity, this is why its concentration must be optimised in certain amplification-based experiments. In this case, the $MgCl_2$ is supplied as a separate vial at 50 mM concentration. This solution must be completely thawed, vigorously vortexed and spun down in a bench-top centrifuge before use.

Reaction conditions

For polymerase-in-one-vial format: resuspend thoroughly using the storage buffer provided (e.g. 250 µl of storage buffer for resuspension of reference UPS54011 at 1 U/µl). Once the resuspension has taken place, handle the polymerase the same way as standard UptiTherm DNA Polymerase (store at –20°C, set up the reaction on ice,tec.).

For aliquoted vials: resuspend thoroughly in 20 µl of bidistilled sterile water. Add your primers, your sample, and complete with water for a final volume of 50 µl. Concentration of reagents (for a final volulme of 50 µl): 200 µM dNTP, 1X buffer, 1 unit polymerase, 2 mM MgCl2 – only for reference UPS54071). Add Mg for reference UPS54081.

Wear disposable gloves and make use of sterile, DNase- and RNase-free pipette tips and tubes in order to avoid contaminations and false negative results.

Notes

*Note 1 : this enzyme is **not** recommended for certain experiments dealing with amplification of sequences homologous to those found in E.coli or very low-annealing temperature amplification approaches (e.g. RAPDs, Random Amplified Polymorphic DNAs).*

Note 2 : at difference with the vast majority of the thermostable DNA polymerases existing in the market, UptiTherm polymerase shows optimal specificity at 2 mM $MgCl_2$ final concentration (rather than 1.5 mM) in reaction buffer.

Notice to buyers/users :

Some of the applications which may be performed with this product are covered by applicable patents in certain countries.

The purchase of this product does not include or provide a license to perform patented applications.

Users may be required to obtain a license depending on the country and/or application.

Associated products and documents

BY1740 GelRed Nucleic Acid Gel Stain 10000X in water
31272L Agarose, regular uses
S54811 100bp DNA ladder