

# DreamTaq DNA polymerases

Thermo Scientific™ DreamTaq™ DNA polymerases are available in standard and hot-start varieties and multiple formats for everyday PCR. Choose DreamTaq Hot Start DNA Polymerase for higher specificity, sensitivity, and yield compared to the standard DreamTaq DNA Polymerase. DreamTaq Hot Start DNA Polymerase also allows for room-temperature reaction setup and reaction stability.

## General recommendations for PCR setup

### Template DNA

- The optimal amount of template DNA for a 50 µL reaction is 0.01–1.0 ng for both plasmid and phage DNA, and 0.1–1.0 µg for genomic DNA.

### Primers

- The recommended concentration range for the PCR primers is 0.1–1.0 µM.
- For degenerate primers and primers used for long-range PCR, use higher primer concentrations in the range of 0.3–1.0 µM.

### MgCl<sub>2</sub> concentration

- 10X DreamTaq Buffer and DreamTaq™ PCR Master Mixes include MgCl<sub>2</sub> for a final reaction concentration of 2.0 mM, which is optimal for the majority of PCR reactions.
- If the DNA samples contain EDTA or other metal chelators, the Mg<sup>2+</sup> concentration should be increased accordingly (one molecule of EDTA binds one Mg<sup>2+</sup> ion).

## Did you choose the right format for your workflow?

Format	DreamTaq DNA Polymerase	DreamTaq Hot Start DNA Polymerase
Stand-alone enzyme—flexibility in PCR setup	Colorless (Cat. No. EP0701/EP0702)*	Colorless (Cat. No. EP1701/EP1702)*
	Green** (Cat. No. EP0711/EP0712)*	Green** (Cat. No. EP1711/EP1712)*
DreamTaq PCR Master Mix—convenience with ready-to-use mix	Colorless (Cat. No. K1071/K1072)	Colorless (Cat. No. K9011/K9012)
	Green** (Cat. No. K1081/K1082)	Green** (Cat. No. K9021/K9022)

\* Additional product sizes available.

\*\* Green gel loading dye already added.

### Reaction setup

1. Gently vortex and briefly centrifuge all solutions after thawing.
2. Place a thin-walled PCR tube on ice for a reaction using standard DreamTaq DNA Polymerase, or mix and add the components, as shown in the table, for each 50  $\mu$ L reaction. Reaction setup on ice is not required for DreamTaq Hot Start DNA Polymerase or master mix.
3. Gently vortex the samples and spin down.
4. When using a thermal cycler that does not have a heated lid, overlay the reaction with 25  $\mu$ L of mineral oil.
5. Perform PCR using the recommended thermal cycling conditions outlined below.

Components	DreamTaq/ DreamTaq Hot Start DNA Polymerase	DreamTaq/ DreamTaq Hot Start PCR Master Mix (2X)
10X DreamTaq Buffer*	5 $\mu$ L	—
dNTP Mix, 2 mM (Cat. No. R0241)	5 $\mu$ L (0.2 mM of each)	—
Forward primer	0.1–1.0 $\mu$ M	0.1–1.0 $\mu$ M
Reverse primer	0.1–1.0 $\mu$ M	0.1–1.0 $\mu$ M
Template DNA	10 pg–1 $\mu$ g	10 pg–1 $\mu$ g
DreamTaq DNA Polymerase/DreamTaq PCR Master Mix (2X)	1.25 U	25 $\mu$ L
Water, nuclease-free	To 50 $\mu$ L total	To 50 $\mu$ L total
<b>Total volume</b>	<b>50 <math>\mu</math>L</b>	<b>50 <math>\mu</math>L</b>

\* 10X DreamTaq Buffer contains 20 mM  $MgCl_2$ , which is optimal for most applications. A final  $MgCl_2$  concentration of 2.0 mM is generally ideal for PCR. If additional optimization is required, the  $MgCl_2$  concentration can be further increased up to 4.0 mM with addition of 25 mM  $MgCl_2$  (Cat. No. R0971). The volume of water should be reduced accordingly.

### Tips for optimizing cycling conditions

- For GC-rich DNA templates, the denaturation step can be prolonged to 3–4 min. DNA denaturation can also be enhanced by the addition of 5–10% glycerol, 5% DMSO, 1% formamide, or 1.0–1.5 M betaine.
- For amplification of templates >6 kb, we recommend reducing the extension temperature to 68°C.
- If the PCR product will be cloned into a TA vector such as in the Thermo Scientific™ InsTAclone™ PCR Cloning Kit (Cat. No. K1213), the final extension step may be prolonged to 15 min to enable the complete 3' dA tailing of the PCR product.

### Cycling conditions

Step	DreamTaq DNA Polymerase/ Master Mix		DreamTaq Hot Start DNA Polymerase/ Master Mix		Number of cycles
	Temp.	Time	Temp.	Time	
Initial denaturation	95°C	1–3 min	95°C	1–3 min	1
Denaturation	95°C	30 sec	95°C	30 sec	25–40
Annealing	$T_m - 5^\circ C$	30 sec	$T_m^*$	30 sec	
Extension**	72°C	1 min	72°C	1 min	
Final extension	72°C	5–15 min	72°C	5–15 min	1

\* Depends on the primer  $T_m$  values. Use the  $T_m$  calculator at [thermofisher.com/tmcalculator](http://thermofisher.com/tmcalculator)

\*\* The recommended extension step is 1 min for PCR products up to 2 kb. For longer products, the extension time should be prolonged by 1 min/kb.

Find out more at [thermofisher.com/dreamtaq](http://thermofisher.com/dreamtaq)