



# TaqDog Hot Start DNA Polymerase

## Products

CAT. #	Description	Volume
HTQDG010	TaqDog Hot Start DNA Polymerase with 10x Reaction Buffer (no dNTP), 1000U	1,000 Units (200 µL)
HTQDG050	TaqDog Hot Start DNA Polymerase with 10x Reaction Buffer (no dNTP), 5000U	5,000 Units (1.0 mL)
HTQDG100	TaqDog Hot Start DNA Polymerase with 10x Reaction Buffer (no dNTP), 10,000U	10,000 Units (2 x 1.0 mL)
HTQDG500	TaqDog Hot Start DNA Polymerase with 10x Reaction Buffer (no dNTP), 50,000U	50,000 Units (10 x 1.0 mL)

## Product Information

**TaqDog Hot Start DNA Polymerase** generates a consistent PCR product across a range of DNA templates, including mouse or human, plasmid and bisulfite converted DNA. The Taq polymerase leaves an adenine overhang at the five-prime end making the PCR product ideal for TA cloning.

*Extension:* 3,000 base-pair extension has been confirmed.

*Speed:* Extension speeds up to 1 Kbp / minute.

**TaqDog Hot start Taq DNA Polymerase** contains an aptamer based Hot Start technology that prevents amplification during PCR setup and minimizes the amplification of primer-dimer and nonspecific products. Initial incubation at 94-95 °C is NOT required. **Note:** The aptamer contains a *three-prime cap* that prevents its amplification, cloning into TA vectors and interference with all conventional downstream analysis. Depending on the amount of Taq used, as well as staining sensitivity, aptamers may be visible as a faint 50 base-pair band when imaged.

## Product Components

	50,000 U	10,000 U	5000 U	1,000 U
Taq Polymerase	10 x 1 ml	2 x 1 ml	1 x 1 ml	1 x 200 µl
10X PCR Buffer	5 x 50 ml	1 x 50 ml	1 x 50 ml	4 x 10 ml
MGSO <sub>4</sub>	2 x 50 ml	1 x 50 ml	1 x 10 ml	2 x 1.5 ml
GC - Extender	3 x 50 ml	3 x 10 ml	2 x 10 ml	2 x 1.5 ml

**Taq Storage Buffer:** (50 mM Tris-HCl (pH 8.0), 100 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 1 % Triton X-100, 50 % glycerol)

**10X PCR Buffer:** 100 mM Tris-HCl (pH 8.8), 100 mM KCl, 100 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 1% Triton X-100

**All components are to be stored at -20 °C**

## Protocol

### 1. Add following components to a PCR tube on ice:

*Note: DNA amount, primer amount and GC Extender use/volume should be optimized.*

Component	50 µl reaction
Template DNA (up to 500 ng)	up to 10 µl
10 µM Forward Primer	0.5 µl
10 µM Reverse Primer	0.5 µl
10X PCR Buffer	5 µl
50 mM MgSO <sub>4</sub>	1.5 µl
10 mM dNTP mix	1 µl
Taq DNA Polymerase	0.2 µl
GC-Extender (Optional)*	5 µl – 15 µl
Nuclease-free H <sub>2</sub> O	up to 50 µl

\*GC-Extender is intended for difficult to amplify templates with GC > 60 %, or for templates longer than 2.5 Kbp. Volume to use (between 5 µl – 15 µl) should be optimized for each template and primer combination.

### 2. Mix contents (either through pipetting multiple times or briefly vortexing).

### 3. Centrifuge briefly and proceed to PCR amplification steps.

### 4. PCR amplification:

*Note: Number of cycles, Annealing temperature and extension should be optimized for each template and primer set.*

Stage	Step	Temperature (°C)	Time	Cycles
1	Denature*	94	10 sec - 30 sec	20-30
	Anneal	55 - 60	10 sec - 30 sec	
	Extend	72	30 sec	
2	Final extension step	72	7 min	1
3	Holding step	4	∞	1

\*No additional high temperature incubation is required for “hot start” activation.

### 5. Product is ready for downstream analysis. For long-term storage store at -20C.

END OF PROTOCOL