

Wonder Taq

Thermostable DNA polymerase

Cat.: EME020001

For research use only

Cat.	Size	Description	Volume
EME020001	1x 1000 units	Wonder Taq	1 x 200µl
		5X Wonder Taq Reaction buffer	6 x 1.5 ml

Storage and stability

2 years at -20°C. Refer to the expiry date printed on the label. Please avoid repeated freeze/thaw cycles.

Wonder Taq is a recombinant thermostable DNA polymerase engineered to give robust amplification and high yield with different PCR templates. Wonder Taq DNA Polymerase is supplied with an optimized 5X Wonder Taq Reaction buffer already containing dNTPs, MgCl₂ and enhancers.

Unit definition

One unit is defined as the amount of enzyme that incorporates 10 nmoles of dNTPs into acid-insoluble form in 30 minutes at 72°C.

5X Reaction buffer

5X Wonder Taq Reaction buffer is an "high performance buffer" (proprietary formulation) obtained through rigorous optimization of each component concentration. 5X Wonder Taq Reaction buffer contains 5 mM dNTPs, 15 mM MgCl₂ stabilizers and enhancers. We don't recommend the addition of other PCR enhancers.

Concentration: 5 units/µl.

Application Protocol

The optimal amplification conditions of Wonder Taq, depend on the system used and have to be determined individually. Optimal enzyme concentration ranges from 1.25-5 units/assay (for 50 µl reactions).

PCR reaction must be set-up on ice.

PCR REACTION CONDITIONS

Briefly centrifuge all reagents before beginning the procedure.

Prepare an amplification mixture by adding reagents to a sterile PCR tube:

Reagent Volume Final Concentration

Reagent	Volume	Final Concentration
5X Wonder Taq Reaction buffer	10 µl	1X
DNA template	variable	variable
primer fwd 20 µM	1 µl	0.4 µM
primer rev. 20 µM	1 µl	0.4 µM
Wonder Taq	0.25 – 1 µl	1.25-5 units
sterile redistilled H ₂ O	Variable (up to 50 µl)	
Final Volume	50 µl	

Amplification parameters are greatly dependent on the template primers and amplification apparatus used.

We suggest these conditions in the first instance:

Cycle step	Temperature (°C)	Time	Cycles
Initial Denaturation	95°C	1 min	1
Denaturation	95°C	15 sec	
Annealing	variable	15 sec	25-35
Extension	72°C	10-30sec/kb	

IMPORTANT NOTES

Primers

The final concentration of primers is usually 0.2-0.6 μ M. Please note that too high primer concentration may cause mispriming and amplification of non specific products.

Template

The recommended amount of template per 50 μ l PCR reaction are listed below:

- For plasmid DNA use 50 pg-10 ng of DNA
- For genomic DNA start with 200 ng. The range should be between 5-500 ng

Avoid using template resuspended in EDTA containing buffers (for example TE) to avoid free Mg^{2+} chelation.

Initial Denaturation

1 min at 95°C is the recommended initial denaturation for plasmid DNA or cDNA.

Longer initial denaturation step (up to 3 min) is required for more complex substrates such as eukaryotic genomic DNA.

Denaturation

For templates with low GC content (40-45%) the cycling denaturation step at 95°C can be decreased to 5 sec.

Annealing

The optimal annealing temperature is usually 2-5°C below the lower T_m of the pair of primers used. Running a temperature gradient is the best way to determine the optimal annealing temperature. 55°C can be used as a starting point. Depending on the reaction the annealing time can be reduce to 5 sec.

Extension

The extension time depends on the length and the complexity of the template:

- for low complexity template (plasmid DNA) with amplicons up to 5 kb 10 sec is sufficient
- for high complexity templates (eukaryotic genomic DNA) over 1 kb we recommend to use longer extension times by incrementing 30 sec for kb.