

# PowerAmp 2X PCRmix-Green

Master Mixes standardise your results



**Cat.No.** RT008G

1250ul Ready to Use supplied as a 2X PCR Mastermix for optimal 25 µl .reactions (100 x 25 µl )

Store at -20°C

Recombinant	○
5' to 3' Exonuclease	○
3' to 5'Exonuclease	✗
Terminal dA Addition	○
Endonuclease Free	○

**Description :** Hot start Taq DNA Polymerase for qPCR is designed for Real-Time PCR and Hot-start PCR. A special inhibition the reaction at room temperature until after the first denaturation step. This prevents primer-dimers and other artefacts.

PowerAmp 2X PCRmix-Green is optimized mixture contain of Hot Start Taq enzyme, reaction buffer, dNTP, enhancer as 2-fold concentration. PowerAmp 2X PCRmix-Green is designed to allow the user for quick , easy preparation of reaction mixture. The PowerAmp 2X PCRmix-Green can be amplification PCR products up to 5 kb and the products can be directly cloning into T-vector.

**storage conditions:** long time at -20°C short time at 4 °C

**Template:** PowerAmp 2X PCRmix-Green is suitable for amplifying targets up to 3 kb from the following templates.

**Genomic DNA:** 10–200 ng

**Plasmid DNA :** 1–5 ng

**cDNA :** ~100 ng starting total RNA

**Primers:** Use 0.3 µM per primer as a general starting point. For larger amounts of template (e.g., 200 ng genomic DNA), increasing the concentration up to 0.5 µM per primer may improve yield.

**Annealing Temperature:** The annealing temperature is slightly higher than with typical PCR. The optimal annealing temperature should be ~2°C lower than the T<sub>m</sub> of the primers used. A range of 58–68°C is recommended.

**Extension Time:** As little as 30 seconds per kb is suitable for most targets. Use up to 60 seconds per kb for maximum yield.

## PCR Protocol:

1. Thaw the 2x Hot start mix at room temperature. Vortex the 2x Hot start mix and then spin it briefly in a micro centrifuge to collect the material in the bottom of the tube.

2. Prepare one of the following reaction mixes on ice:

Component	Volume
2x Hotstart mix	12.5 ul
Primer1 (20 pmol)	1-2 ul
Primer2 (20 pmol)	1-2 ul
template	1-5 ul
ddH <sub>2</sub> O	Up to 25 ul
Total	25 ul

3. If necessary you can scale up your volume

## Program the thermal cycler as follows

Step	Temperature	Time	Cycle
Initial denaturation	94-95°C	5-10mins	1
Denaturation	94-95°C	0.2-2mins	20-40
Annealing	50-68°C	0.2-2mins	
Extension	72°C	1min/1kb	
Final extension	72°C	1-10mins	1

## Step

After cycling, maintain the reaction at 4°C. Samples can be stored at -20°C until use.

Analyze products using standard agarose gel electrophoresis.

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