PCR SuperMix

Cat. No.: SM200-0100 Size: 100 Reactions (4 X 1.125ml)
Cat. No.: SM200-0025 Size: 25 Reactions (1 X 1.125ml)
Store at -20°C

Description

PCR SuperMix provides qualified reagents for the amplification of nucleic acid templates by the polymerase chain reaction (PCR). PCR SuperMix contains Mg++, dNTPs, and recombinant Taq DNA Polymerase at concentrations sufficient to allow amplification during PCR. PCR SuperMix is supplied at 1.1X concentration to allow approximately 10% of the final reaction volume to be used for the addition of primer and template solutions. Reagents sufficient for 100 amplification reactions of 50 µl each are provided.
PCR SuperMix may be stored at either -20°C or 4°C. Storage at 4°C avoids the necessity of thawing the mix before assembling the PCR. No detectable reduction of PCR performance or enzyme activity is observed after storage of PCR SuperMix for 12 months at 4°C. Repeated freeze-thaw cycles do not reduce performance or activity.

Components

22 mM Tris-HCl (pH 8.4), 55 mM KCl, 1.65 mM $MgCl_2$, 220 µM dGTP, 220 µM dATP, 220 µM dTTP, 220 µM dCTP, and stabilizers.
This product is distributed for laboratory research only. CAUTION: Not for diagnostic use. The safety and efficacy of this product in diagnostic or other clinical uses has not been established.

Quality Control

PCR SuperMix is evaluated in a DNA polymerization activity assay that measures the percent of Taq DNA polymerase inhibition versus an uninhibited control.
A functional assay is also performed. Components of PCR SuperMix are tested for the absence of DNase, RNase and exonuclease activities.
Recombinant Taq DNA polymerase is tested for the absence of exonuclease, and double- and single-stranded endonuclease activities. The enzyme is >90% homogeneous as determined by SDSpolyacrylamide gel electrophoresis.

Guidelines and Recommendations

Since PCR is a powerful technique capable of amplifying trace amounts of DNA, all appropriate precautions should be taken to avoid crosscontamination.Ideally, amplification reactions should be assembled in a DNA-free environment. Use of aerosol-resistant barrier tips is recommended. Take care to avoid contamination of PCR SuperMix with the primers or template DNA used in individual reactions.
PCR products should be analyzed in an area separate from the reaction assembly area.A standard 50-µl reaction uses 45 µl of PCR SuperMix and 5 µl of primer and template solutions. For the primer sets used in the development of PCR SuperMix, no decrease in product yield was observed if the amount of template and primer solution added is between a fraction of a microliter and 20 µl. Lower yield occurs as the Mg++ concentration drops to a suboptimal level. If the final Mg++ concentration is adjusted to 1.5 mM, the volume of primer and template solution that can be added to 45 µl of PCR SuperMix can exceed 50 µl.

Protocol

The following protocol is suggested as a starting point and guideline when using PCR SuperMix. We recommend assembling reactions on ice from pre-chilled components. This protocol is for a reaction size of approximately 50 µl. The reaction size may be adjusted as desired.
Note: For multiple reactions with common components, prepare a master mix of the components common to all reactions to reduce pipetting errors.

1. Set up reaction tubes/plates on ice.
2. Add the following components in any order to each reaction vessel.
   - 45-µl PCR SuperMix
   - Primers (200 nM final concentration per primer is recommended)*
   - Template DNA solution*

   * The total volume of primer and template solution should be 0.5–20 µL. If the final Mg concentration is adjusted to 1.5 mM, the volume of primer and template solution that can be added to 45 µL of PCR SuperMix can exceed 50 µL.
3. Mix contents and cover with mineral or silicone coil, if necessary.
4. Cap reaction vessels and then briefly centrifuge the contents.
5. Process in the thermal cycler for 25~35 cycles as follows:

Initial Denaturation	2 minutes at 94°C	
Denaturation	15 seconds at 94°C	25-35 cycles
Annealing	30 seconds at the proper annealing temperature	
Extension	1 mins / kb at 72°C	
	Hold at 4 °C	