

PCR-Master Mix

DESCRIPTION

The 2x PCR Master Mix contains all reagents required for PCR and is designed to make PCR as easy and simple as possible. All components (inclusive Taq DNA-Polymerase respectively Hot Start DNA Polymerase) are provided in an optimized concentration in the 2x PCR-Master solution. With 2x PCR Master Mix all you need to do is to add primers and template DNA, thus minimizing the pipetting effort and possible sources of error. The mix is suitable for PCR amplification of DNA-fragments up to 4 kb long, in most cases even longer targets can be successfully amplified.

Red PCR Master Mix and Red Hot Start PCR Master Mix contain an inert red dye which allows online monitoring of the electrophoresis. The dye has no adverse effect on PCR.

QUALITY CONTROL ASSAYS

Functional Assays	Specification
4 kb PCR	suitable
Physical Assays	Specification
Endonuclease Assay	no activity
DNase Assay	no activity
RNase Assay	no activity

BASIC PROTOCOL

Combine the following components in a PCR-reaction tube and adjust to a final volume of 50 µl with H₂O:

Component	Volume	Final concentration
2 x PCR-Master Mix	25 µl	1 x (1.5 mM MgCl ₂)
Primer A	variable	0.2 - 1 µM
Primer B	variable	0.2 - 1 µM
Template DNA	variable	1 - 150 ng
H ₂ O	variable	
Final volume	50 µl	

Mix well, centrifuge and place in thermal cycler. Optimal conditions for concentration of primer, template and temperature profile need to be determined for each reaction.

TROUBLESHOOTING

Observation	Check
Nonspecific products (smearing)	<ul style="list-style-type: none">- Concentration of enzyme, primer, and/or dNTPs was too high- Annealing temperature for primers too low- Too many cycles- Annealing and extension time too long- Too much template DNA
Low yield of product	<ul style="list-style-type: none">- Concentration of enzyme, primer, and/or dNTPs was too high- Annealing temperature for primers too low- Too many cycles- Annealing and extension time too long- Too much template DNA
No product	<ul style="list-style-type: none">- Incorrect annealing temperature- Incomplete denaturation- Poorly designed primers- Use of destroyed components due to wrong storage

STORAGE

Store at -20°C. Multiple freeze-thaw-cycles should be avoided by preparing aliquots.

WARNING

For research use only. Not recommended or intended for diagnosis of disease in humans or animals. Do not use internally or externally in humans or animals.