PCR Master Kit, 2x

Cat. No.: MM2001
Store: -20°C

Quantity: 400 Reactions
Shipment: Wet Ice

Description:
The PCR Master offers convenient reagents for PCR amplifications. The reagent of Master Mix is an optimized ready to use 2X PCR mixture of Taq DNA Polymerase (recombinant), PCR buffer, $MgCl_2$ and dNTPs. Master Mix contains all components for PCR, except DNA template and primers. Additionally, sterile water and mineral oil, PCR grade is supplied. The supplied PCR Master Kit is sufficient for 400 amplification reactions of 25 µl volume each or 200 amplification reactions of 50 µl volume.

The kit offers convenient solutions for amplification of up to 5 kb obviating individual adjustment of the reagent compositions. PCR products generated by PCR Master Kit resemble 3` single A-over-hang products.

Components (supplied):

Master Mix	4 × 1250µl
Control DNA (4pg/µl)	200µl
Forward Primer	40µl
Reverse primer	40µl
Distilled Water	5ml
Mineral Oil	4ml

Stability:
The kit is stable at -20°C for 24 months. Repeated freezing and thawing should be avoided.

General Protocol for DNA amplification:
The PCR Master can be used for nearly all PCR applications. The only limitation is that the sample volume must not exceed half the total reaction volume. The optimal reaction conditions (incubation temperatures and times, concentration of template DNA and primer) depend on the template/primers system and must be determined individually.

All solutions should be thawed on ice, gently vortexed and briefly centrifuged. Add in a thin walled PCR tube on ice:

*For a total 25µl reaction volume

Component of a sample	Volume	Final concentration
Master Mix	12.5µl	1X
Forward Primer	Variable	0.1-1µM
Reverse Primer	Variable	0.1-1µM
Template DNA	Variable	10pg-1µg
Sterile Deionized Water	To 25µl	-

Note: - annealing temperature depends on the melting temperature of the primer used.
- Elongation time depends on fragment length. We recommend e.g. 45- 120 sec for 1.5 –5 kb.
- Elongation temperature depends on the length of amplification product: 72°C are used for amplifications up to 3 kb:
68°C are used for amplifications > 3 kb.

Cycling parameters for positive control and control primers (Included):

Initial Denaturation	95°C	120 Sec	
Denaturation	93°C	10-45 Sec	**35 cycles**
Annealing	40-65°C	10-60 Sec	
Extension	72°C	20-90 Sec	

Result analysis for positive control and control primers (Included):
Analyze 10 µl of amplified samples in a 1% agarose gel after adding loading buffer.
The presence of 620 bp fragment indicates of positive.
For gel electrophoresis use of 100 bp Ladder (Cat. No.: SL7031) is recommended.

For Research Use Only

SinaClon BioScience

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