

Hot Start Master

Master mix of thermally activated DNA polymerase for high specificity

Ready-to-use mixes for PCR

Cat.-No.	Amount	Size
PCR-103S	100 reactions	1 ml
PCR-103L	500 reactions	5 ml

For *in vitro* use only

Quality guaranteed for 12 months

Store at -20°C, avoid frequent thawing and freezing

Storage at 4°C for up to 3 months possible

Description

Hot Start Master is recommended for high specificity PCR reactions and shows superior amplification at lowest template concentrations. It contains all reagents required for PCR (except template and primer) in a premixed 5x concentrated ready-to-use solution. The thermal activation prevents the extension of nonspecifically annealed primers and primer-dimers formed at low temperatures during PCR setup. The polymerase mix contains monoclonal antibodies which block polymerase activity prior to the onset of thermal cycling.

Activation step

Hot Start Pol requires no prolonged heating or denaturing step. The antibodies are quickly inactivated at the increased temperature of thermal cycling.

Recommended PCR assay

50 µl PCR assay		
10 µl	5x Hot Start Master Mix	red cap
0.2-1 µM	each Primer	
2-50 ng	Template DNA	
Fill up to 50 µl	PCR grade H ₂ O	white cap

Recommended cycling conditions

Activation step and initial denaturation	94°C	2 min	1x
Denaturation	94°C	30 sec	30x
Annealing ¹⁾	45 - 68°C	30 sec	
Elongation ²⁾	72°C	30 sec - 3 min	
Final elongation	72°C	2 min	1x

- 1) The annealing temperature depends on the melting temperature of the primers used.
- 2) The elongation time depends on the length of the fragments to be amplified. A time of 1 min/kbp is recommended.

For optimal specificity and amplification an individual optimization of the recommended parameters may be necessary for each new template DNA and/or primer pair.

5x Hot Start Master (red cap)

Thermally activated DNA polymerase, dATP, dCTP, dGTP, dTTP, reaction buffer with KCl and MgCl₂, stabilizers

PCR grade water (white cap)