

Hot Start Master Lyophilisate

Lyophilized hot start master mix

Ready-to-use lyophilisates for PCR

Cat. No.	Amount	Size
PCR-153S-8TS	12 strips / 96 reactions	8-tube strips
PCR-153L-8TS	60 strips / 480 reactions	
PCR-153S-FTP	2 plates / 192 reactions	96-well plates (flat top, without skirt)
PCR-153L-FTP	10 plates / 960 reactions	
PCR-153S-HSP	2 plates / 192 reactions	96-well plates (half skirt)
PCR-153L-HSP	10 plates / 960 reactions	

For *in vitro* use only

Quality guaranteed for 12 months

Store below 25°C

Store in an aluminium-coated bag or on a dry place

Lyophilisates may hydrate at humidity levels >70%

when sealing is opened

Hot Start Master Lyophilisate

Preloaded lyophilisates of hot start DNA polymerase, dATP, dCTP, dGTP, dTTP, reaction buffer with MgCl₂, and stabilizers

PCR-grade water

Description

Hot Start Master Lyophilisate is delivered in PCR reaction tube strips or 96-well plates preloaded with a complete hot start master mix in a dry, room temperature stable format. The lyophilisate combines highest performance with convenience of use and stability. There is no need for freezing, thawing or pipetting on ice. The few remaining pipetting steps minimize the risk of errors or contaminations.

Each vial contains polymerase, dNTPs and reaction buffer with MgCl₂ required for a 20 µl PCR assay.

To perform PCR, fill up the vials with a premix of primers and PCR-grade water and add DNA template. If necessary, centrifuge to remove bubbles, vortex the vials to assure homogeneity and start cycling.

The Hot Start Master Lyophilisate provides improved specificity and sensitivity when amplifying low-copy-number targets in complex backgrounds. The polymerase activity is blocked at ambient temperature and switched on automatically at the onset of the initial denaturation. The thermal activation prevents the extension of nonspecifically annealed primers and primer-dimers formed during PCR setup.

Activation step

Hot Start Master Lyophilisate requires no prolonged heating or denaturing step. The polymerase inhibiting ligand is quickly released at the increased temperature of thermal cycling.

Recommended PCR assay

20 µl PCR assay		
forward Primer	0.2-1 µM	0.4-2 µl / 10 µM
reverse Primer	0.2-1 µM	0.4-2 µl / 10 µM
Template DNA	1-50 ng	
PCR grade H ₂ O	fill up to 20 µl	

Recommended cycling conditions

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 primer-template pair.