Data Sheet

IFTA AG
Certified QMS according to
DIN EN ISO 9001
Reg. No. IC 03214 034

Jena Bioscience

Multiplex PCR Master Lyophilisate

Lyophilized Master Mix for multiplex PCR

Ready-to-use lyophilisates for PCR

	Cat. No.	Amount	Size
	PCR-161S-8TS	12 strips / 96 reactions	8-tube strips
	PCR-161L-8TS	60 strips / 480 reactions	
	PCR-161S-FTP	2 plates / 192 reactions	96-well plates (flat top, without skirt)
	PCR-161L-FTP	10 plates / 960 reactions	
	PCR-161S-HSP	2 plates / 192 reactions	96-well plates (half skirt)
	PCR-161L-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

Multiplex PCR Master Lyophilisate
Preloaded lyophilisates of Multiplex PCR Mastermix containing Hot Start Taq polymerase, nucleotides, optimized reaction buffer, stabilizers

PCR-grade water (white cap)

Description

Multiplex PCR Master Lyophilisate is delivered in PCR reaction tube strips or 96-well plates preloaded with a complete 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.
To perform PCR, fill up the vials with 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 lyophilisate is specially designed for the set-up of multiplex PCR reactions. It contains an optimized composition of polymerase, nucleotides, $MgCl_2$ and stabilizing components in a specifically developed buffer system allowing the parallel amplification of a multitude of fragments in a single PCR assay.
The lyophilisate is recommended for use in routine PCR reactions and highly suitable for multiple target gene amplification in a single tube.
The high specificity and sensitivity of the mix is achieved by an optimized hot-start polymerase. Its activity is blocked at ambient temperature preventing the extension of nonspecifically annealed primers and primer-dimer formations at low temperatures during PCR setup.

Recommended PCR assay

Prepare a primer mix to reduce pipetting errors. Pipet with sterile filter tips and perform the setup in an area separate from DNA preparation or analysis. No-template controls should be included in all amplifications.

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component	stock conc.	final conc.	20 µl assay
Multiplex PCR Master Lyophilisate		1x	1 tube
forward primer 1	10 µM	400 nM	0.8 µl
reverse primer 1	10 µM	400 nM	0.8 µl
forward primer 2	10 µM	400 nM	0.8 µl
reverse primer 2	10 µM	400 nM	0.8 µl
forward primer …	10 µM	400 nM	0.8 µl
reverse primer …	10 µM	400 nM	0.8 µl
Template - animal genomic DNA - bacterial genomic DNA - plasmid and lambda DNA			10-200 ng 1-50 ng 1-5 ng
PCR-grade water			fill up to 20 µl

Recommended cycling conditions

	temperature	time	cycles
initial denaturation	95°C	10 min	1x
denaturation	95°C	30 sec	30-50x [2]
annealing [1]	58-64°C	40 sec	
elongation [3]	72°C	1 min/kb	
final elongation	72°C	5 min	1x

[1] The optimal annealing temperature (AT) can be calculated for each primer as following:
AT = T_m - 5°C with T_m = 2°C·(A+T) + 4°C·(G+C)
Please note that primers should be designed to show minimal differences in there melting temperatures (T_m).

[2] Cycle numbers are recommended as following:
- animal genomic DNA
  - 10-50 ng: 35-50 cycles
  - 50-200 ng: 30-45 cycles
- bacterial genomic DNA
  - 1-5 ng: 35-50 cycles
  - 5-50 ng: 30-40 cycles
- plasmid and lambda DNA
  - 1-5 ng: 30-40 cycles

[3] The elongation time depends on the length of the fragments to be amplified. A time of 1 min per kb is recommended.

For optimal specificity and amplification an individual optimization of the recommended parameters may be necessary for each new primer-template combination.