DATA SHEET

Hot Start Taq Pol Master Mix (2X) Green

Master mix of Hot Start Taq Polymerase for direct gel loading

	Cat. Nº.	Amount
☐	POL-117XS	50 reactions
☐	POL-117S	100 reactions
☐	POL-117M	200 reactions
■	POL-117L	500 reactions
☐	POL-117XL	1.000 reactions

Shipping:
Shipped on blue ice

Storage Conditions:
Store at -20 °C

Additional Storage Conditions:
Avoid freeze/thaw cycles. Hot Start Taq Pol Master Mix (2X) is also stable for three months at 4°C, so for frequent use, an aliquot may be kept at 4°C.

Shelf Life:
12 months

For *in vitro* use only!

Description:
Hot Start Taq Pol Master Mix contains Hot Start Taq Polymerase in an optimized PCR buffer with Mg^{2+} and dNTPs. The master mix is supplemented with tracking dyes for direct loading of PCR products on gels. It contains all reagents required for PCR (except template and primer) in a premixed 2x concentrated ready-to-use solution. The Master Mix is recommended for use in routine PCR reactions. It is optimized for high specificity and guarantees minimal by-product formation. Antibody-based hot start technology avoids nonspecific amplification and enables room temperature reaction setup. The tracking dyes in the master mix do not interfere with PCR performance and are compatible with downstream applications such as fluorescent automatic DNA sequencing, ligation, and restriction digestion. The blue dye migrates with 3-5 kb fragments and the yellow dye migrates faster than 20 bp (1% agarose gel).

Kit contents:
2x Hot Start Taq Pol Master Mix Green (purple cap)
Master mix of thermostable Hot Start DNA polymerase, dATP, dCTP, dGTP, dTTP, KCl, $MgCl_2$, dye, gel loading buffer and stabilizers.

PCR Reaction Setup

Use the quantities below to prepare a single 50 µl PCR reaction. Thaw, mix, and briefly centrifuge each component before use. Add the following components to a microcentrifuge tube:

1. Prepare PCR master mix

Note: Consider the volumes for all components listed next steps to determine the correct amount of water required to reach your final reaction volume.

Components	50 µL rxn	[final]
Water, grade PCR	To 50 µl	
2 X Hot Start Taq Pol Master Mix	25 µl	1X

Mix and briefly centrifuge the components.

2. Add template DNA and primers

Components	50 µL rxn	[final]
Foward primer (10 µM)	0,5 - 2,5 µl	0,1 – 0,5 µM
Reverse primer (10 µM)	0,5 - 2,5 µl	0,1 – 0,5 µM
DNA template		10 pg – 1 µg**

**genomic DNA: 1 ng-1µg; plasmidial ou viral DNA: 1 pg-1 ng

Cap each tube, mix, and briefly centrifuge the content.

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3. Incubate reactions in a thermal cycler.

Recommended cycling conditions:

Step		Temp.	Time
Initial denaturation		95 ºC	1 min
30 cycles	Denaturation	95 ºC	15 - 30 sec
	Annealing [1]	45-68 ºC	15 - 30 sec
	Elongation [2]	72 ºC	30 sec - 4 min
Final extension		72 ºC	2 min
Hold		4 - 8 ºC	

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/kb 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.