DATA SHEET

cellco

Taq Pol High Fidelity Hot Start

Heat activated DNA polymerase for high accuracy *and specificityThermus* and *Pyrococcus* species, recombinant, *E. coli*

Cat. Nº.	Amount
☐ POL-136XS	250 units
■ POL-136S	500 units
☐ POL-136M	1.000 units

Unit Definition:
One unit is defined as the amount of the enzyme required to catalyze the incorporation of 10 nmoles of dNTPs into an acid-insoluble form in 30 minutes at 74 °C.

Concentration:
2,5 units/µL

Shipping:
Shipped on blue ice

Storage Conditions:
Store at -20 °C

For *in vitro* use only!

Additional Storage Conditions:
Avoid freeze/thaw cycles

Shelf Life:
12 months

Kit contents:

Taq High Fidelity Hot Start Pol (blue cap)
2,5 units/µl Taq High Fidelity Hot Start Pol in storage buffer.

Taq High Fidelity Pol Reaction Buffer complete (red cap) - 10x

Description:
High Fidelity Hot Start Pol is based on a blend of Taq DNA polymerase and a proofreading enzyme specially designed for highly accurate and efficient amplification. The additional hot-start function provides improved specificity and sensitivity when amplifying lowcopy-number targets in complex backgrounds or when prolonged room-temperature set up is required. 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-dimer formation at low temperatures during PCR setup. The enzyme shows excellent results with extremely long (up to 20 kb), GC-rich or other difficult templates. The enzyme blend includes a highly processive 5′ 3′ DNA polymerase and possesses a 5′ 3′ polymerization-dependent exonuclease replacement activity. Its inherent 3′ 5′ exonuclease proofreading activity results in a greatly increased fidelity of DNA synthesis compared to Taq polymerase. The enzyme is highly purified and free of bacterial DNA.

Activation step:
High Fidelity Hot Start Pol requires no prolonged heating or denaturing step. The polymerase inhibiting antibodies are released at the increased temperature of the initial denaturation.

Fidelity of the enzyme:
High Fidelity Pol is characterized by a 4-fold higher fidelity compared to Taq polymerase.
$ER_{HighFidelityPol} = 3.4 \times 10^{-6}$
The error rate (ER) of a PCR reaction is calculated using the equation
ER = MF/(bp x d), where MF is the mutation frequency, bp is the number of base pairs of the fragment and d is the number of doublings
(2^d = amount of product / amount of template).

Content:

High Fidelity Hot Start Pol (blue cap)
2.5 units/µl High Fidelity Hot Start Polymerase in storage buffer

High Fidelity Buffer (red cap)
10x conc.

Recommended 50 µl PCR assay

Components	50 µL rxn	[final]
10x HF/HS Reaction Buffer	5 µl	1x
dNTP (Mix 10 mM)	1 µl	200 µM
each primer	0,5 - 2,5 µl	0,2 - 0,5 µM
Taq HF/HS Pol (2,5U/ µl)	0,5µl	1,25 U/reaction
DNA template	-	10 pg - 1 µg
Water, grade PCR	fill up to 50 µl	

Please note that it is essential to add the polymerase as last component.

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Incubate reactions in a thermal cycler.

Recommended cycling conditions:

Step		Temp.	Time
Initial denaturation		95 °C	2 min
20 - 30 cycles	Denaturation	95 °C	20 sec
	Annealing [1]	50 - 68 °C	30 sec
	Elongation [2]	68 °C	1 min/kb
Final elongation		68 °C	1 min/kb
Hold		4 - 8 °C	

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.