Cyrusbioscience

2X Ready to Load PCR Master Mix

Cat. Number: 21010

Storage: Store at -20°C.

Package: 1.25mL/vial, 4 vials/set

Composition:

0.2 U/µl Taq DNA polymerase, Loading Dye, Reaction Buffer, 3 mM MgCl2, 0.4 mM of each dNTP (dATP, dCTP, dGTP and dTTP).

Sources: *Thermus aquaticus*

Error Rate: $1/10^5$

Application

Polymerase Chain Reaction (PCR)
DNA labeling reactions
Sequencing/cycle sequencing
Primer extension

Description

Taq DNA Polymerase was originally isolated from the thermophilic eubacterium Thermus aquaticus, a strain lacking Taq I restriction endonuclease. Taq catalyzes the polymerization of nucleotides into duplex DNA in the 5'→3' direction in the presence of magnesium. The enzyme has an apparent molecular weight of 94,000 daltons by SDS-PAGE and exhibits 5'→3' exonuclease activity, lacks 3'→5' exonuclease activity. Taq is recommended for use in PCR and primer extension reactions at elevated temperature.

2X Ready to Load PCR Master Mixis a ready-to-use 2X solution containing Taq DNA polymerase, dNTPs, MgCl2 and reaction buffers at optimal concentrations for PCR. Simply add primers, template, and water to amplify the target sequence and other molecular biology applications. This formulation not only saves valuable time, but also reduces number of pipetting and reagent handling errors.

Taq DNA Polymerase has the independent terminal transferal activity which results in the addition of a single nucleotide (adenosine) at 3' end of the extension product. TA cloning vector is recommended if the extension product is needed to be cloned.

Recommended Basic PCR Usage:

1. Gently vortex and briefly centrifuge 2X Ready to Load PCR Master Mix after thawing.
2. Set up each reaction as follows:

Component	50 µl reaction	Final Concentration
2X Ready to Load PCR Master Mix	25µl	1x
Forward Primer	Variable	0.1–1 µM
Reverse Primer	Variable	0.1–1 µM
Template DNA	variable	<1µg
ddH2O	to 50µl	

3. Gently mix the solution a few times and spin down.

4. Perform PCR using the recommended thermal cycling conditions outlined below:

(For maximum yield and specificity, temperatures and cycling times should be optimized for each new template target or primer pair)

STEP	TEMP	TIME
Initial Denaturation	95°C	30sec
30 Cycles	95°C 45-68°C 72°C	15-30sec 15-60sec 1min/kb
Final Extension	72°C	5min
Hold	4°C	

CERTIFICATE OF ANALYSIS

Endonuclease Assay

No conversion of covalently closed circular DNA to nicked form was detected after incubation of 1X Taq DNA Polymerase Mastermix with 1 µg of supercoiled plasmid DNA (pUC19) in for 4 hours at 37°C.

Exonuclease Assay

No degradation of DNA was observed after incubation of 1 µg of lambda DNA/HindIII fragments in 1X Taq DNA Polymerase Mastermix for 4 hours at 37°C.

Functional Assay

Good performance in PCR was tested for amplification of 1.8 kb gene.