Taq DNA Polymerase

10X Standard Mg^{2+} Free Buffer

5 units/µl

Cat. No.	Units	10X Mg^{2+} Free Buffer	$MgCl_2$ 25 mM
AO112203	500	1.5 mL	1.5 mL
AO112204	1,000	2 x 1.5 mL	2 x 1.5 mL
AO112206	2,500	4 x 1.5 mL	4 x 1.5 mL
AO112207	5,000	3 x 5 mL	3 x 5.0 mL
AO112208	10,000	6 x 5.0 mL	6 x 5.0 mL
AO112209	20,000	8 x 5.0 mL	8 x 5.0 mL

Store at -20°C. *For in-vitro laboratory use only*

General Description

AS ONE Taq DNA Polymerase is a thermostable recombinant DNA polymerase, which exhibits very high activity in primer extension and other molecular biology applications. The enzyme is isolated from *Thermus aquaticus* and has a molecular weight of approximately 94 kDa.

AS ONE Taq DNA Polymerase has both a 5'→3' DNA polymerase and a 5'→3' exonuclease activity. The enzyme lacks a 3'→5' exonuclease activity (no proofreading ability). Taq DNA Polymerase leaves an A′ overhang, which makes the enzyme ideal for TA cloning.

The 10X Reaction Buffer provided does not contain Mg^{+2}. 25 mM $MgCl_2$ is supplied separately.

10X Mg^{++} Free Standard Buffer

100 mM Tris-HCl pH 8.5, 500 mM KCl, 1% Triton X-100.

Unit Definition

One unit is defined as the amount that incorporates 10 nmoles of dNTPs into acid-precipitable form in 30 minutes at 72°C under standard assay conditions.

Storage and Dilution Buffer

Enzyme is supplied in 20 mM Tris-HCl pH 8.3, 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, 0.5% Tween 20®, 0.5% NP40, 50% glycerol.

Quality Control

Each lot of Taq DNA Polymerase is tested for contaminating activities, with no trace of endonuclease activity, nicking activity, exonuclease activity or priming activity.

Suggested Protocol using Taq DNA Polymerase

This protocol serves as a guideline only. Optimal reaction conditions may vary and must be individually determined.

- In some applications, $MgCl_2$ is needed for the best results. For this reason, 25 mM $MgCl_2$ is included with the kit.

1. Thaw 10X Mg^{2+} Free Standard Buffer, dNTP mix, primer solutions. **It is important to mix the solutions completely before use to avoid localized concentrations of salts**.

2. Prepare a master mix according to Table 1. The master mix typically contains all the components needed for extension except the template DNA.

Table 1. ***Reaction components (master mix & template DNA)***

Component	Vol./reaction	Final Conc.
10X Mg^{2+} Free Buffer	5 µL	1X
$MgCl_2$, 25 mM	1- 9 µL	0.5 – 4.5 mM
dNTP mix (12.5 mM of each)	0.8 µL	0.2 mM of each dNTP
Primer A	Variable	0.1–1.0 µM
Primer B	Variable	0.1–1.0 µM
Taq DNA Polymerase	Variable	1-5 units
Template DNA	Variable	Variable
Distilled Water	Variable	- - - -
TOTAL volume	50 µL	- - - -

Table 2. ***$MgCl_2$ concentration in a 50 µL reaction***

Final $MgCl_2$ conc. in reaction (mM)	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5
Volume of 25 mM $MgCl_2$ per rxn (µL):	1	2	3	4	5	6	7	8	9

Table 2 provides the volume of 25 mM $MgCl_2$ to add to the master mix if a certain $MgCl_2$ concentration is required.

3. Mix the master mix thoroughly and dispense appropriate volumes into reaction tubes. Mix gently, *e.g.*, by pipetting the master mix up and down a few times.

4. Add template DNA to the individual tubes containing the master mix.

5. Program the thermal cycler according to the manufacturer's instructions. For maximum yield and specificity, temperatures and cycling times should be optimized for each new template target or primer pair.

6. Place the tubes in the thermal cycler and start the reaction.