

# EcoTaq HotStart DNA Polymerase, 5U/ $\mu$ L

250U, 1000U

**Cat No:** ETHS025, ETHS100

**Shipping** : Ship with blue ice.

**Storage** : Store at -20°C.

## General Information

EcoTaq HotStart DNA Polymerase is a mixture of a monoclonal anti-Taq antibody and recombinant Taq DNA Polymerase, specifically designed for high-specificity PCR applications. The antibody binds to the Taq DNA polymerase and inhibits its activity at low temperatures, effectively preventing non-specific primer annealing and primer-dimer formation prior to thermal cycling.

During the initial denaturation step of PCR, the antibody is inactivated, thereby restoring full DNA polymerase activity and enabling a true hot start effect. This mechanism improves amplification specificity and yield without requiring additional enzyme activation steps or modifications to standard PCR protocols.

EcoTaq HotStart DNA Polymerase possesses 5'→3' DNA polymerase activity and 5'→3' exonuclease activity, but lacks 3'→5' exonuclease (proofreading) activity. The enzyme exhibits an extension rate of approximately 2 kb/min and is capable of amplifying DNA fragments up to 5 kb in length. PCR products generated with this enzyme contain a single 3'-A overhang, making them suitable for direct T/A cloning.

EcoTaq HotStart DNA Polymerase offers high amplification efficiency and robust performance, making it ideal for routine PCR, DNA sequencing, cloning, and other molecular biology applications requiring enhanced specificity and reliability.

EcoTaq DNA Polymerase is supplied together with 10x Reaction Buffer. 10×PCR Buffer contains 15 mM MgCl<sub>2</sub>.

## PCR Setup

| Component                                   | Amount           |
|---------------------------------------------|------------------|
| EcoTaq HotStart DNA Polymerase, 5U/ $\mu$ l | 1 $\mu$ L        |
| 10x Reaction Buffer                         | 5 $\mu$ l        |
| Forward primer, 10 $\mu$ M                  | 2 $\mu$ l        |
| Reverse primer, 10 $\mu$ M                  | 2 $\mu$ l        |
| dNTP Mix, 10 mM each                        | 1 $\mu$ l        |
| Template DNA                                | <0.5 $\mu$ g     |
| ddH <sub>2</sub> O                          | up to 50 $\mu$ l |
| Total                                       | 50 $\mu$ l       |

**Important Note:** PCR reaction conditions might need optimization with additional MgCl<sub>2</sub> (not provided) depending on the primer binding properties.

**Important Note:** The reaction can be prepared at room temperature, and the reagents must be kept on ice.

## PCR Reaction Condition

| Temperature | Time      | Cycles |
|-------------|-----------|--------|
| 94°C        | 2 min     |        |
| 94°C        | 30 sec    |        |
| 55-65°C     | 30 sec    | 30-35  |
| 72°C        | 30 sec/kb |        |
| 72°C        | 2 min     |        |
| 4°C         | $\infty$  |        |