# Taq DNA Polymerase

**with 10X Reaction Buffer**

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### Product Information

**Catalogue numbers:** PCR001 | PCR002 | SP-PCR001
**Enzyme concentration:** 5 U/µL
**Use:** Routine PCR amplification

### Storage & Handling

Store at -20°C; avoid repeated freeze-thaw cycles.
Thaw Reaction Buffer on ice and mix well before use.
Keep polymerase cold during reaction setup.

## Principle and Applications

Taq DNA Polymerase is a thermostable recombinant DNA polymerase derived from Thermus aquaticus. It extends a primer annealed to a DNA template in the 5′→3′ direction, enabling repeated amplification of defined DNA targets during PCR. The supplied 10X Reaction Buffer provides the reaction environment for routine amplification. The enzyme lacks 3′→5′ proofreading exonuclease activity and can generate 3′-A overhangs suitable for TA cloning.

## Ordering Information and Kit Contents

| Component / pack | PCR001 | PCR002 | Sample pack |
|---|---|---|---|
| **Catalogue number** | PCR001 | PCR002 | SP-PCR001 |
| **Pack size** | 200 U | 1,000 U | 100 U |
| **Taq DNA Polymerase (5 U/µL)** | 40 µL | 200 µL | 20 µL |
| **10X Reaction Buffer** | 1 mL | 3 mL | 1 mL |
| **50 mM $MgCl_2$ solution** | 100 µL | 400 µL | 50 µL |

## Standard PCR Reaction Setup

Prepare reactions on ice using nuclease-free tubes and pipette tips. The following composition is a starting point for routine PCR.

| Component | 25 µL PCR | 50 µL PCR | Final concentration |
|---|---|---|---|
| **DNA template** | Variable | Variable | According to sample |
| **Forward primer (10 µM)** | 0.5 µL | 1.0 µL | 0.2 µM |
| **Reverse primer (10 µM)** | 0.5 µL | 1.0 µL | 0.2 µM |
| **dNTP mix (10 mM each)** | 0.5 µL | 1.0 µL | 200 µM each |
| **10X Reaction Buffer** | 2.5 µL | 5.0 µL | 1X |
| **Taq DNA Polymerase (5 U/µL)** | *0.25 µL | *0.5 µL | 1.25 U / 25 µL; 2.5 U / 50 µL |
| **Nuclease-free water** | To 25 µL | To 50 µL | Final volume |

**$MgCl_2$:** A separate 50 mM $MgCl_2$ solution is included for optimization. Supplementation depends on the magnesium already present in the Reaction Buffer; determine the required amount using the confirmed buffer specification.

* Optimization needed, this is the standard protocol.

## Recommended Thermal Cycling Programme

Example programme for amplification of an approximately 1 kb DNA fragment. Annealing temperature and extension duration depend on primer Tm, amplicon length and template properties.

| Step | Temperature | Duration | Cycles |
|---|---|---|---|
| **Initial denaturation** | 95°C | 5 min | 1 cycle |
| **Denaturation** | 95°C | 30 s | **35 cycles** |
| **Annealing** | 55°C | 30 s | |
| **Extension** | 72°C | 45 s | |
| **Final extension** | 72°C | 7 min | 1 cycle |
| **Hold** | 4°C | Until recovery | — |

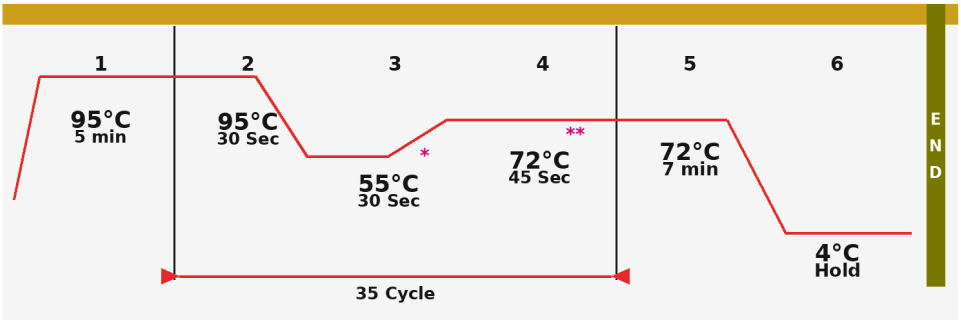


Figure 1. Recommended thermal cycling profile for routine endpoint PCR.

Optimization note. Adjust annealing temperature to suit the primer pair and increase the 72°C extension step for longer amplicons. As a practical starting point, use approximately 1 minute of extension time per kb of target length.

## Representative Amplification Result

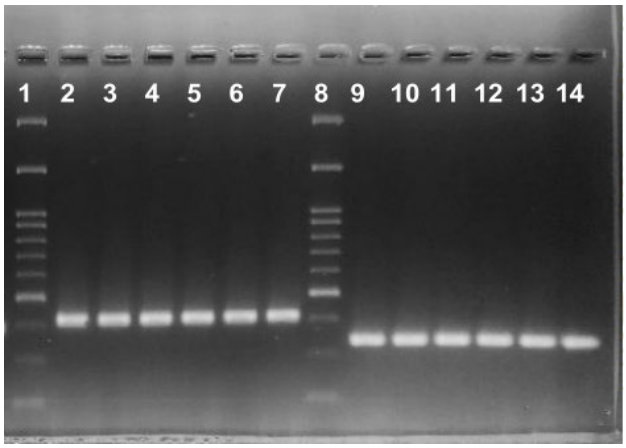


**Figure 2 | Comparative amplification of plant genomic DNA targets**

Representative agarose gel electrophoresis of PCR products amplified from plant genomic DNA.

Lane 1: DNA ladder; lane 2: Vendor A; lanes 3 and 11: G2P Taq DNA Polymerase; lanes 12–14: Vendors B, C and D, respectively.

## Reaction Component Guidance

| Component | Practical guidance |
|---|---|
| **DNA template** | Use clean DNA. Excess template or extraction carryover can cause nonspecific amplification or inhibition. |
| **Primers** | Use a matched forward/reverse primer pair; begin near 0.2 µM each and optimize as needed. |
| **dNTPs** | A typical starting concentration is 200 µM of each dNTP in the final reaction. |
| **$MgCl_2$** | Magnesium affects specificity and yield; adjust using the separate 50 mM stock only when the buffer formulation and target final concentration are known. |

## Troubleshooting Guide

Use the following suggestions if the reaction does not produce the expected product. Optimize one variable at a time, keeping a positive control and a no-template control alongside samples.

| Observation | Possible cause | Recommended action |
|---|---|---|
| **Little or no PCR product** | Low-quality or insufficient template | Check DNA integrity and concentration; repeat purification if inhibition is suspected. |
| **Little or no PCR product** | Primer or annealing mismatch | Verify primer sequences; optimize annealing temperature and primer concentration. |
| **Little or no PCR product** | Enzyme or cycling conditions | Confirm polymerase addition, denaturation, cycle count and final extension. |
| **Multiple bands** | Annealing temperature too low | Increase annealing temperature or use a gradient PCR programme. |
| **Multiple bands / smearing** | Excess template, primers or $Mg^{2+}$ | Reduce input or titrate reagent concentrations; verify $MgCl_2$ content of the buffer. |
| **Primer-dimer bands** | Primer self-complementarity | Review primer design and lower primer concentration when appropriate. |
| **Poor GC-rich amplification** | Difficult secondary structures | Consider a validated GC-rich PCR strategy and adjust denaturation/annealing conditions. |
| **Band in negative control** | DNA or amplicon contamination | Replace suspect reagents, use filtered tips and separate PCR setup from post-PCR analysis. |
| **Inconsistent results** | Variable handling or freeze–thaw | Mix Reaction Buffer after thawing, make a master mix and keep the enzyme cold. |

# Additional Information

**Enzyme unit definition.** One unit of Taq DNA Polymerase is the amount of enzyme required to incorporate 10 nmol of dNTPs into acid-insoluble material in 30 minutes at 72°C using poly(dA)-oligo(dT) as template-primer.

**Storage.** Keep the enzyme at -20°C and avoid repeated freeze–thaw cycles. The stated storage stability is up to one year at -20°C. Return the enzyme to the recommended temperature promptly after use.

**Reaction Buffer and $MgCl_2$.** The kit includes 10X Reaction Buffer and 50 mM $MgCl_2$ solution as separate components. Use the $MgCl_2$ stock only when further magnesium optimization is required, accounting for magnesium already present in the Reaction Buffer.

**Application scope.** For research use only. Product is intended for routine PCR amplification. Taq DNA Polymerase does not have proofreading activity and may not be the appropriate choice when a high-fidelity polymerase is required.

Product information: G2P Taq DNA Polymerase is supplied under catalogue numbers PCR001 and PCR002; sample pack catalogue number is SP-PCR001. For technical assistance, contact G2P using the details provided in this datasheet.