



HAMI HAYAT AYANDEGAN

حامی حیات آیندگان

Red Bio Tech

آدرس:

تهران، صادقیه، بلوار فردوس شرق، خیابان  
ولیعصر، کوچه تقدیری، ساختمان سهند،  
پلاک 16، واحد 18

همراه:

09358888140

تلفن:

02144962980

آدرس وبسایت:

[www.hamihayatco.com](http://www.hamihayatco.com)

ایمیل:

[info@hamihayatco.com](mailto:info@hamihayatco.com)

## 2X MASTER MIX

Name	Polymerase 2x Master Mix
Color	Red
Volume	1 ml
Reaction	100

### Description

As a prepared and ready-to-use solution, 2× Taq Mix consists of Taq DNA Polymerase, dNTPs, Mg<sup>2+</sup> and Reaction Buffer at optimal concentrations for effective amplification of DNA templates by PCR.

### Composition:

- 0/06 U/μl of taq DNA polymerase,
  - 3 mM mgcl<sub>2</sub>
  - 400 μM of each dNTPs.
  - reaction buffer
- It is convenient to prepare the final PCR by only add primers and template DNA.
  - This premixed formulation has high efficiency as takes less time by simplifying the process.
  - It is also reproducible since reduces contamination because of the fewer pipetting steps needed for PCR set-up.
  - The mix retains all features of Taq DNA Polymerase which is a thermostable recombinant DNA polymerase derived from thermophilic bacterium *Thermus aquaticus*.
  - Having 94 kDa molecular weight, Taq DNA Polymerase can amplify DNA target up to 5 kb (simple template), and the elongation velocity is 0.9~1.2kb/min (70~75°C).
  - It has 5' to 3' polymerase activity but lacks of 3' to 5' exonuclease activity that results in a 3'-dA overhangs PCR product.

### Limitation

This product is exclusively for in vitro research purposes and is not applicable in theragnostic for humans or animals.

### Applications

- High throughput PCR
- Routine PCR with high reproducibility
- Generation of PCR products for TA cloning

## Basic PCR Protocol

All solutions should be thawed on ice, gently vortex and briefly centrifuge.

1. Add the following components to a sterile micro centrifuge tube sitting on ice:

Reagent	Quantity	Final concentration
2× Taq Mix	25 µl	1×
Forward Primer	variable	0.4-1 µM
Reverse Primer	variable	0.4-1 µM
Template DNA	variable	10pg-1µg
Water, nuclease-free	to 50 µl	-

Recommendation amounts of template DNA in a 50 µl reaction mix:

Human genomic DNA	0.1µg-1µg
Plasmid DNA	0.5ng-5ng
Phage DNA	0.1ng-10ng
E. coli genomic DNA	10ng-100ng

2. Combine contents in the tube. Cap tubes and centrifuge briefly to collect the contents to the bottom. When using a thermal cycler that does not contain a heated lid, overlay the reaction mixture with 25 µl mineral oil.

3. Conduct 25-35 cycles of PCR amplification as follows:

Initial Denaturation	94°C	3 minutes
25-35 Cycles	94°C	30 seconds
	55-68°C	30 seconds
	72°C	1-10 minutes
Final Extension	72°C	10 minutes

4. Incubate for an additional 10 min at 72°C and maintain the reaction at 4°C. The samples can be stored at -20°C until use.
5. Analyze the amplification products by agarose gel electrophoresis and use nucleic acid dye staining to visualize. Use appropriate molecular weight standards.

## Notes on cycling conditions

- Being suitable for most PCR applications, recombinant Taq DNA Polymerase has the following features:
- The half-life: >40 minutes at 95°C.
- The error rate in PCR:  $2.2 \times 10^{-5}$  errors per nt per cycle
- It accepts modified nucleotides (e.g., biotin-, digoxigenin-, fluorescent-labeled nucleotides) as substrates for the DNA synthesis.
- The number of PCR cycles depends on the amount of template DNA in the reaction mix and on the expected

yield of the PCR product. 25-35 cycles are usually sufficient for the majority PCR reactions. Low amounts of starting template may require 40 cycles.

**Recommendations to lower the risk of contamination are as follows:**

- Prepare your DNA sample, set up the PCR mixture, perform thermal cycling and analyze PCR products in separate areas.
- Set up PCR mixtures in a laminar flow cabinet equipped with an UV lamp.
- Wear fresh gloves for DNA purification and reaction set-up.
- Use reagent containers dedicated for PCR. Use positive displacement pipettes, or use pipette tips with aerosol filters to prepare DNA samples and perform PCR set-up.
- Always perform “no template control” (NTC) reactions to check for contamination.

## Quality Control

**High Fidelity and High speed:** The fidelity is 6 times of the regular Taq. And the extension speed is reach to 5 sec/kb

**Super stability:** The special premixed receipt makes the mixture stable at least 28 days under room temperature (RT).

**High efficiency:** A new type of high-efficiency polymerase, with strong amplification ability, can easily amplify various types of templates.

Compared with normal PCR mix, the time of pre denaturation and denaturation is very short, and the extension speed can reach to 5 secs/kb! Greatly save PCR reaction time!

The result shows the Super master mix is very stable, can stable at room temperature (+10°C\_ +15°C) at least 28 days.