

# 2.0X Taq Master Mix, Clear (1.5mM MgCl<sub>2</sub>) Cat #: 42-134B

Contents: 1000 Reactions

Storage: -20°C.

Reagent for in vitro laboratory use only

## **General Description**

Apex Taq DNA Polymerase Master Mix, Clear from Genesee Scientific is a ready-to-use 2.0X reaction mix. Simply add primers, template, and water to successfully carry out primer extensions and other molecular biology applications.

Apex Taq DNA Polymerase, an optimized NH<sub>4</sub><sup>+</sup> buffer system, dNTPs and magnesium chloride are present in the Tag DNA Polymerase Master Mix, Clear. Each reaction requires 25 µl of the 2.0X reaction mix. Simply add primers, template and water to a total reaction volume of 50 µl.

Taq DNA Polymerase Master Mix, Clear offers several advantages. Optimized for increased specificity of DNA targets up to 4 kb. Setup time is significantly reduced. The chance of contaminating component stocks is eliminated. Reduction of reagent handling steps leads to better reproducibility. Standard tests can be set up with the confidence that results will be consistent every time.

# Composition of the Taq Master Mix, Clear (2.0X)

- Tris-HCl pH 8.5, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 3 mM MgCl<sub>2</sub>, 0.2 % Tween® 20
- 0.4 mM of each dNTP
- Apex Taq DNA polymerase

# Storage and Stability

Long term storage at -20 °C. Product expiry at -20 °C is stated on the label.

Option: Store at +4 °C for up to 6 months.

### **Quality Control**

Tag DNA Polymerase is tested for contaminating activities, with no traces of endonuclease activity, nicking activity or exonuclease activity.

#### Protocol

This protocol serves as a guideline for primer extensions. Optimal reaction conditions such as incubation times, temperatures, and amount of template DNA may vary and must be individually determined.

#### Notes:

- Set up reaction mixtures in an area separate from that used for DNA preparation or product analysis.
- The table below shows the reaction set up for a final volume of 50 µL.
- Keep all components on ice.
- **Important:** Mix the solutions completely before use to avoid localized concentrations of salts.
  - 1. Set up each reaction as follows:

Table 1.

Component	Vol./Reaction	Final Conc.
2.0X Taq Master Mix, Clear	25 μL	1X
Primer A	Variable	0.1–1.0 μΜ
Primer B	Variable	0.1–1.0 μΜ
Nuclease-Free Water	Variable	
Template DNA	Variable	Variable
TOTAL volume	50 μL	

- 2. Mix gently by pipetting the solution up and down a few times.
- 1. Program the thermal cycler according to the manufacturer's instructions and recommendations in table 2. (DNA targets < 1kb) or table 3. (DNA targets 1 – 4 kb)

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

Table 2. Three-step PCR program for targets < 1kb

Cycles	Duration of cycle	Temperature
1	2-5 minutes	95 °C
25-35	20 – 30 seconds <sup>a</sup> 20-40 seconds <sup>b</sup> 30 seconds <sup>c</sup>	95 °C 55 – 60 °C 72 °C
1	5 minutes <sup>d</sup>	72 °C



Table 3. Three-step PCR program for targets 1kb-4kb

Cycles	Duration of cycle	Temperature
1	2-5 minutes	95 °C
25-35	20 – 30 seconds <sup>a</sup> 20-40 seconds <sup>b</sup> 60 – 300 seconds <sup>c</sup>	95 °C 55 – 60 °C 72 °C
1	5 minutes <sup>d</sup>	72 °C

- a. Denaturation step: This step is the first regular cycling event and consists of heating the reaction to 95 °C for 20 – 30 seconds. It causes melting of the DNA template by disrupting the hydrogen bonds between complementary bases, yielding single-stranded DNA molecules.
- $^{\text{b.}}$  Annealing step: The reaction temperature is lowered to  $50-65\,^{\circ}\text{C}$  for 20-40 seconds allowing annealing of the primers to the single-stranded DNA template. Typically, the annealing temperature is about  $3-5\,^{\circ}\text{C}$  below the  $T_m$  (melting temperature) of the primers used.
- c. Extension/elongation step: Taq polymerase has its optimum activity temperature at 72 °C. At this step the DNA polymerase synthesizes a new DNA strand complementary to the DNA template strand. The extension time depends on the length of the DNA fragment to be amplified. As a rule of thumb, at its optimum temperature the DNA polymerase will polymerize a thousand bases per minute.
- d. Final elongation: This single step is occasionally performed at a temperature of 72 °C for 5 minutes after the last PCR cycle to ensure that any remaining single-stranded DNA is fully extended.

#### **Related Products**

Taq Polymerase kits (500 units)	Cat#
With 10X Standard and Ammonium Reaction Buffer	42-800B1
With 10X Combination Buffer	42-800B3
Glycerol Free	42-800B4

Hot Start DNA Polymerase (500 units)	Cat#
With 10X Ammonium and Combination Reaction Buffer	42-106

All polymerases are also available in kits,  $Mg^{2+}$  free buffers and 50 mM  $MgCl_2$ .

Master Mixes (500 reactions)	Cat#
2X Taq RED Master Mix, 1.5 mM MgCl <sub>2</sub>	42-138
2X Taq Master Mix, Clear, 1.5 mM MgCl <sub>2</sub>	42-134
2X Hot Start Master Mix Buffer I, 1.5 mM MgCl <sub>2</sub>	42-198
2X Hot Start Master Mix Buffer I Blue, 1.5 mM MgCl <sub>2</sub>	42-144

The shown Hot Start master mixes are ammonium based. Also available with balanced ammonium and potassium based buffers.

Real-time PCR (400 reactions)	Cat#
qPCR 2X Master Mix for Probe, without $ROX^{TM}$	42-116P
qPCR 2X Master Mix for Probe, low ROX™	42-118P
qPCR 2X Master Mix for Probe, high ROX™	42-120P
qPCR 2X GREEN Master Mix, without ROX™	42-116PG
qPCR 2X GREEN Master Mix, low ROX™	42-118PG
qPCR 2X GREEN Master Mix, high ROX™	42-120PG

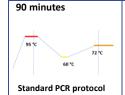
DNA Ladders	Cat#
Apex 100 bp-Low DNA Ladder, 250 applications	19-109
Apex 1 kb DNA Ladder, 333 applications	19-115
Apex 200 bp DNA Ladder, 200 applications	19-111
ECON Mini DNA Ladder 100-500 bp, 100 applications	19-130
ECON Low DNA Ladder 100-1000 bp, 100 applications	19-131
ECON PCR Ladder 100-3000 bp, 100 applications	19-132
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Ultrapure dNTPs	Cat#
dNTP set, 100 mM each: 250 μl of each dA, dC, dG and dT	42-410
dNTP Set, 100 mM each: 1 ml of each dA, dC, dG and dT	42-403
dNTP Mix 40 mM (1 x 500 μl): 10 mM each dA, dC, dG, dT	42-411
dNTP Mix 100 mM (2 x 1 ml): 25 mM each dA, dC, dG, dT	42-405
dNTP Mix 10 mM (10 x 1 ml): 2.5 mM each dA, dC, dG, dT	42-406

Other concentrations and Single dNTPs are available

Accessory reagents	Cat#
50 mM MgCl <sub>2</sub> , 3 × 1.5 ml	42-303
Nuclease-Free Water, PCR Grade, 6 x 5 ml	42-710

# Nuclease-Free Water, PCR Grade, 6 x 5 ml 42-710 Inspiration for Fast PCR Fast PCR protocol: Apex Taq DNA Polymerases and Taq DNA master mixes



SAVE 1 HOUR

PCR program	for 3-step	Standard Po	CR
Cycler step	Temp.	Duration	Cycles
Initial heating	95 °C	3 min.	1
Denaturation	95 °C	30 sec.	
Annealing*	60 °C	30 sec.	30
Extension	72 °C	30 sec.	
Final extension	72 °C	5 min.	1

31 minutes
98 °C 72 °C 72 °C 60 °C

- just chang	ge PCR cy	cler settings	!
PCR progran	for 2-ste	p Fast PCR	
Cyclor stop	Tomp	Duration	

Cycler step	Temp.	Duration	Cycles
Initial heating	98 °C	40 sec.	1
Denaturation	92 °C	2 sec.	
Extension*	60 °C	2 sec.	30
Final	72 °C	5 min.	1
extension	72 C		

2-step Fast PCR protocol

\* the extension temperature depends on the primer set. For fast PCR choose highest possible T<sub>rop</sub>

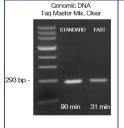
A DNA
Taq DNA Polymerase
STANDARD FAST

300 bp 90 min 31 min

# Amplification of λ DNA -Taq DNA Polymerase

Reaction mix*	
Ammonium buf.	1x
dNTP mix	0,2 mM each
MgCl <sub>2</sub>	1,5 mM
Primers	0,2 μΜ
λDNA	1 ng
Taq DNA pol.	0.5 – 1U

\* H₂O up to a total volume of 25 μl



Amplification of gDNA -	
2x Taq Master Mix, Clea	r

Reaction mix*	
Taq MM Clear	1x
Primers	0,2 μΜ
gDNA	20 ng

<sup>\*</sup> H₂O up to a total volume of 25 μl