



# Apex Hot Start 2.0X Master Mix Blue

with Apex Buffer I

**Cat #: 42-143**

**2.0X Master Mix Kit (1.5mM Final MgCl<sub>2</sub> Conc.)**

**Unit size:** 100

**Contents:** 2 x 1.25ml

**Storage:** -20°C.

Reagent for *in vitro* laboratory use only

## General Description

Apex Hot Start Master Mix BLUE is a ready-to-use 2.0X master mix. Simply add primers, template and water to successfully carry out primer extensions and other molecular biology applications.

Apex Hot Start DNA Polymerase, NH<sub>4</sub><sup>+</sup> buffer system, dNTPs and magnesium chloride are present in Apex Hot Start Master Mix with Apex Buffer I. 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.

Apex Hot Start DNA Polymerase is a modified form of Apex Taq DNA Polymerase, which is activated by heat treatment. A chemical moiety is attached to the enzyme at the active site, which renders the enzyme inactive at room temperature. Thus, during setup and the first ramp of thermal cycling, the enzyme is not active and misprimed primers are not extended. The result is higher specificity and greater yields when compared to standard DNA polymerases.

Apex Hot Start Master Mix BLUE offers several advantages: Direct gel loading, no need to use separate loading dyes for electrophoresis and subsequent visualization, and 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 2.0 X Apex Hot Start Master Mix BLUE w/ buffer I

- 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 dNTPs
- Apex Hot Start DNA Polymerase
- Inert Blue Dye
- Stabilizer

## 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.
- **Important:** Mix the solutions completely before use to avoid localized concentrations of salts.

1. Set up each reaction as follows:

Component	Vol./Reaction	Final Conc.
Apex Hot Start Master Mix Blue with Apex Buffer I	25 µL	1X
Primer A	Variable	0.1–1.0 µM
Primer B	Variable	0.1–1.0 µM
PCR Grade Water	Variable	----
Template DNA	Variable	Variable
<b>TOTAL volume</b>	50 µL	----

2. Mix gently by pipetting the solution up and down a few times.
3. Program the thermal cycler according to the manufacturer's instructions.
4. **Each program must start with an initial heat activation step at 95°C for 15 minutes.**

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

A typical thermal cycling program is shown below:

<b>95°C for 15 min.</b>		Activate Apex Hot Start Polymerase
<b>30-40 cycles:</b>		
<b>95°C</b>	30 sec.	Denature template
<b>45-65°C</b>	30 sec.	Anneal primer
<b>72°C</b>	1-5 min.	Elongation
<b>72°C for 5 min.</b>		Elongation

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

## 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

High Fidelity - Proof reading (500 units)	Cat#
Hi-Fi PR™ Taq 2.5 U/μl	42-110

All polymerases are also available in kits, Mg<sup>2+</sup> free buffers and 50 mM MgCl<sub>2</sub>.

Master Mixes (500 reactions)	Cat#
2X Taq DNA Polymerase Master Mix, 1.5 mM MgCl <sub>2</sub>	42-132
2X Taq RED Master Mix, 1.5 mM MgCl <sub>2</sub>	42-138
2X Hot Start Master Mix Buffer I, 1.5 mM MgCl <sub>2</sub>	42-198

The shown 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™	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

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.

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
Apex ECON Mini DNA Ladder, 100 applications	19-130
Apex ECON Low DNA Ladder, 100 applications	19-131
Apex ECON PCR Ladder, 100 applications	19-132

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

Tween 20® is a registered trademark of ICI Americas, Inc.