



*******SAMPLE PACK*******

Apex Hot Start 2.0X Master Mix Blue

with Apex Buffer I

Cat #: SMP42-144

2.0X Master Mix Kit (1.5mM Final MgCl₂ Conc.)

Unit size: 20 Reactions

Contents: 1 x 0.5 ml

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, NH₄⁺ 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, (NH₄)₂SO₄, 3 mM MgCl₂, 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 |
| TOTAL volume | 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:

| | | |
|-------------------------|----------|------------------------------------|
| 95°C for 15 min. | | Activate Apex Hot Start Polymerase |
| 30-40 cycles: | | |
| 95°C | 30 sec. | Denature template |
| 45-65°C | 30 sec. | Anneal primer |
| 72°C | 1-5 min. | Elongation |
| 72°C for 5 min. | | 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, Mg²⁺ free buffers and 50 mM MgCl₂.

| Master Mixes (500 reactions) | Cat# |
|--|--------|
| 2X Taq DNA Polymerase Master Mix, 1.5 mM MgCl ₂ | 42-132 |
| 2X Taq RED Master Mix, 1.5 mM MgCl ₂ | 42-138 |
| 2X Hot Start Master Mix Buffer I, 1.5 mM MgCl ₂ | 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 ₂ , 3 x 1.5 ml | 42-303 |
| Nuclease-Free Water, PCR Grade, 6 x 5 ml | 42-710 |