

RT² SYBR Green / ROX qPCR Master Mix

<u>Catalog Number</u>	<u>Description</u>
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PA-012	For 2 RT ² Profiler™ PCR Arrays OR 200 25- μ l reactions (2.5 ml)
PA-012-12	For 12 RT ² Profiler PCR Arrays
PA-012-24	For 24 RT ² Profiler PCR Arrays
PA-112	2000 25- μ l reactions (25 ml)

Description

The RT² SYBR Green / ROX qPCR master mix contains all of the reagents and buffers required for real-time polymerase chain reactions in ABI® and Stratagene® instrumentation: real-time PCR buffer, a high-performance HotStart DNA Taq polymerase, nucleotides, SYBR® Green dye, and the ROX reference dye needed to normalize the instruments' optics. Simply add the master mix to PCR tubes along with your template and primers. The chemically-modified and tightly controlled HotStart enzyme uniquely provides more accurate SYBR Green results by preventing the amplification of primer dimers and other non-specific products. This qPCR master mix is best suited for real-time PCR applications using SYBR® Green based detection on ABI® and Stratagene® instrumentation.

Contents

Please check the kit components immediately after you receive this package. SuperArray is not responsible for any missing items not reported within two (2) business days upon receipt.

PA-012:	Two (2) tubes each containing 1.4 ml of 2X solution and enough for 100 standard 25- μ l reactions
PA-012-12:	Twelve (12) tubes each containing 1.4 ml of 2X solution and enough for one PCR Array
PA-012-24:	Twenty-four (24) tubes each containing 1.4 ml of 2X solution and enough for one PCR Array
PA-112:	One (1) bottle containing 25 ml of 2X solution and enough for 2000 standard 25- μ l reactions

Storage Conditions

The kit is shipped on dry ice or cold packs. For long-term storage, keep at -20 °C. If entire volume is not be used all at once, divide into aliquots and store at -20 °C. Avoid repeated freezing and thawing.

Shelf Life

All reagents are stable for 6 months after receipt if stored at the recommended temperature.

Product Specification Sheet

Recommendations for Use

1. Ensure that you do not contaminate the 2X real-time PCR cocktail by dividing into aliquots containing the amount of cocktail necessary for the number of reactions you are preparing each day. The rest of the cocktail should be kept in storage away from any sources of template DNA.
2. When using RT² qPCR Master Mixes with RT² Profiler™ PCR Arrays, RT² qPCR Primer Assays or CHIP-qPCR Primer Assays, refer to the appropriate User Manuals.
3. For general real-time PCR purposes, mix the following components in a PCR tube:
 - 12.5 µl RT² SYBR Green / ROX qPCR Master Mix
 - 10.5 µl ddH₂O
 - 1.0 µl template cDNA (up to 250 ng)
 - 1.0 µl gene-specific 10 µM PCR primer pair stock
 - 25.0 µl final volume
4. Recommended real-time thermal cycler program:
NOTE: *The 10 min step at 95 °C is required to activate the HotStart Taq DNA polymerase.*
 - 95 °C, 10 min;** 40 cycles of (95 °C, 15 sec; and 60 °C, 60 sec)
5. Program the real-time thermal cycler to detect and record the SYBR® Green signal from every reaction at the end of the 60 °C annealing / extension step of each cycle.
6. Run your instrument's default melting curve program immediately after the above PCR program, and generate a first derivative dissociation curve. No more than one peak should appear in each reaction

If your instrument does not have a default melting curve program, run the following program instead:

95 °C, 1 min; 65 °C, 2 min (OPTICS OFF); 65 °C to 95 °C at 2 °C / min (OPTICS ON).

NOTE: *Be sure to visually inspect the plate after the run for any signs of evaporation from any of the wells. If evaporation is observed, make a note of which wells so that you may qualify your data analysis appropriately.*

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