Quality Control Procedures:

Screening for nuclease and protease contamination, as well as functional testing of the Master Mix has been completed prior to lot release.

For further details or Certificate of Conformance contact Launchworks with the lot number of the Master Mix.

The information in this guide is subject to change without notice.

DISCLAIMER: TO THE EXTENT ALLOWED BY LAW, LAUNCHWORKS AND/OR ITS AFFILIATE(S) WILL NOT BE LIABLE FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING YOUR USE OF IT.





Instructions for Use

One-Step RT-qPCR Master Mix (No passive dye) Thomas Number: CHM01P929

Note: Follow safe chemical handling protocols. Wear appropriate protective eyewear, clothing, and gloves.

Intended Use and Product Description:

The One-Step Master Mix (No passive dye) is used to perform one-step single or multiplexed real time PCR applications with any gene-specific primer and probe sets and is suitable for both RNA and DNA targets.

Isolate and purify the target nucleic acid samples according to your laboratory practices. Contact Launchworks for sample preparation recommendations.

Contents and Storage:

10 x 1mL 2X One-Step RT-qPCR Master Mix

Avoid excess freeze-thaw cycles on each tube of Master Mix.

Store Master Mix at or below -20°C per expiration dating on label. Do not heat, incubate, or keep at ambient temperature. Keep mix on ice while setting up RT-PCR assays.

Protocol:

- 1. Thaw all reagents and keep on ice until set-up is complete.
- Add the following to one well per sample. Calculate the total volume required for each reaction based on the following table.

| Component | Standard System Set-Up (20uL/reaction) | Notes | |
|--|--|---|--|
| RNAse-Free Water | 4 μL | Should be RT-PCR Grade Nuclease-Free Water. | |
| Primer and Probe Set (20X – see note) | 1 μL | Recommended final primer concentrations of 400-600nM and a probe concentration of 200-250nM. | |
| 1-Step Master Mix (2X) | 10 μL | Ensure 2X Master Mix is thoroughly mixed in each well. | |
| RNA Sample or Control | 5 μL | This may be adjusted if needed, by also adjusting the volume of water. Do not exceed 20uL total per reaction. | |
| Total Volume Per Reaction | 20 µL | Mix contents of each reaction thoroughly, once all components are present. Centrifugation of plates or tubes for 3 min at 2,000g (4°C) is recommended to avoid bubbles. | |

| 3. | RT-real-time PCR | Thermal | Cycling | Conditions: |
|----|------------------|---------|---------|-------------|
|----|------------------|---------|---------|-------------|

| Step | Cycles | Temperature | Time | |
|-----------------------|--------|-------------|----------------------------------|--|
| Reverse Transcription | 1 | 53 °C | Hold 10 mins. (ramp rate 4.4) | |
| Denaturation | 1 | 95 °C | Hold 2 mins. (ramp rate 4.4) | |
| American | 45 | 95 °C | Hold 15 sec. (ramp rate 4.4) | |
| Amplification | | 60 °C | Hold 30 sec. (ramp rate 2.2) | |
| Cooling | 1 | 40 °C | Hold 30 sec. (ramp rate 2.2) | |

Note: Data acquisition during 60°C hold of Amplification phase. Ramp rates may vary depending on PCR equipment.

Shelf Life:

Internal shelf-life testing is currently ongoing and is currently set at 12 months when stored at or below -20 $^\circ C.$

Warnings and Limitations:

This material is sold as Research Use Only and should be used in a laboratory setting by trained and qualified professionals only. While we make every effort to ensure the safety of our products, we recommend handling any biological materials with standard precautions as if capable of spreading infectious disease. We do not recommend use of the product if it has passed its expiration date, or if there is evidence of leakage.



ThomasSci.com 833.544.SHIP (7447)

Connect With Us:



CustomerService@ThomasSci.com