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# TaqMan® Fast Advanced Master Mix Product Insert

Insert PN 4444602 Rev. C

## Product part numbers and storage conditions

| Product | Quantity/part number | | | | | | Storage conditions |
|---|---|---|---|---|---|---|---|
| | 1 × 1 mL | 1 × 5 mL | 2 × 5 mL | 5 × 5 mL | 10 × 5 mL | 1 × 50 mL | |
| TaqMan® Fast Advanced Master Mix | 4444556 | 4444557 | 4444963 | 4444964 | 4444965 | 4444558 | Store at -15 to -25 °C until first use, then store at 4 °C. |

## Protocol for TaqMan® and Custom TaqMan® Gene Expression Assays

**Note:**

This Product Insert briefly describes how to perform gene expression experiments using the TaqMan® Fast Advanced Master Mix with TaqMan® and Custom TaqMan® Gene Expression Assays. For more detailed procedures, or for procedures on performing gene expression experiments with TaqMan® MicroRNA Assays or TaqMan® Array Micro Fluidic Cards, refer to the *TaqMan® Fast Advanced Master Mix Protocol* (PN 4444605).

### Prepare the PCR reaction mix

1. Thoroughly mix the TaqMan® Fast Advanced Master Mix.
2. Thaw frozen samples and frozen TaqMan® assays on ice. Resuspend by vortexing, then briefly centrifuge.
3. Calculate the total volume required for each component: *volume for 1 reaction* × *the total number of reactions*.

| Component | Volume (µL) for 1 reaction | | Final concentration |
|---|---|---|---|
| | 384-well plate | 96-well and 48-well plates (both Standard and Fast) | |
| TaqMan® Fast Advanced Master Mix (2×) | 5.0 | 10.0 | 1× |
| TaqMan® Gene Expression Assay (20×) or Custom TaqMan® Gene Expression Assay (20×) | 0.5 | 1.0 | 1× |
| cDNA template | 1.0 | 2.0 | 100 ng to 1 pg |
| Nuclease-free water | 3.5 | 7.0 | |
| **Total volume per reaction** | **10.0** | **20.0** | – |

4. Add all components to a 1.5-mL microcentrifuge tube, cap the tube, then vortex briefly to mix.
5. Centrifuge the tube briefly to spin down the contents and eliminate air bubbles.

### Prepare and run the PCR reaction plate

1. Transfer the appropriate volume of PCR reaction mix to each well of an optical reaction plate.
2. Cover the reaction plate, then centrifuge the plate briefly to spin down the contents and eliminate air bubbles.
3. Run the PCR reaction plate using the parameters below.
   - Run mode:

| Applied Biosystems Real-Time PCR System | Mode |
|---|---|
| 7900HT, 7900HT Fast (384-Well Block Module and Standard 96-Well Block Module), 7500, and 7300 systems | Standard |
| ViiA™ 7, StepOne™, StepOnePlus™, 7900HT Fast (Fast 96-Well Block Module), and 7500 Fast systems | Fast |

- Thermal cycling conditions:

| Applied Biosystems Real-Time PCR System | Thermal-cycling profile | | | | |
|---|---|---|---|---|---|
| | Parameter | UNG incubation† | Polymerase activation‡ | PCR (40 cycles) | |
| | | Hold | Hold | Denature | Anneal/ extend |
| | Temp. (°C) | 50 | 95 | 95 | 60 |
| 7900HT system | Time (mm:ss) | 02:00 | 00:20 | 00:01 | 00:20 |
| 7900HT Fast system (Fast 96-Well, Standard 96-Well, or 384-Well Block Modules) | | | | | |
| ViiA™ 7 system | | | | | |
| StepOne™ system | | | | | |
| StepOnePlus™ system | | | | | |
| 7500 Fast system | Time (mm:ss) | 02:00 | 00:20 | 00:03 | 00:30 |
| 7500 system | | | | | |
| 7300 system | | | | | |

† Required for optimal UNG activity.
‡ Required to activate the AmpliTaq® Fast DNA Polymerase.

### Analyze the results

Data analysis varies, depending on the instrument. For further information, refer to the *TaqMan® Fast Advanced Master Mix Protocol* (PN 4444605) and the appropriate documentation for your instrument.

## Safety information

**IMPORTANT!**
For safety and biohazard guidelines, refer to the "Safety" section in the *TaqMan® Fast Advanced Master Mix Protocol* (PN 4444605).

### Obtaining SDSs

The Safety Data Sheet (SDS) for any chemical supplied by Applied Biosystems is available to you free 24 hours a day. To obtain SDSs:

1. Go to **www.appliedbiosystems.com**, click **Support**, then select **SDS**.
2. In the Keyword Search field, enter the chemical name, product name, SDS part number, or other information that appears in the SDS of interest. Select the language of your choice, then click **Search**.
3. Find the document of interest, right-click the document title, then select any of the following:
   - **Open** – To view the document
   - **Print Target** – To print the document
   - **Save Target As** – To download a PDF version of the document to a destination that you choose

**Note:**
For the SDSs of chemicals not distributed by Applied Biosystems, contact the chemical manufacturer.



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