

# TaqMan® Fast Virus 1-Step Master Mix


Insert PN 4444464 Rev. A

For Research Use Only. Not intended for any animal or human therapeutic or diagnostic use.

## Perform the RT-PCRs for the target sample

### Prepare the samples

Isolate and purify the target nucleic acid samples according to your laboratory practices. See the *TaqMan® Fast Virus 1-Step Master Mix Protocol* (PN 4453800) for some sample preparation recommendations.

 **Note:** You can use this protocol for both RNA and DNA targets. During thermal cycling, the reverse transcription step will not affect performance with DNA targets.

### Prepare the reactions in a reaction plate

1. Thaw all reagents on ice.
2. Gently invert the TaqMan® Fast Virus 1-Step Master Mix 2 to 3 times to ensure a homogenous solution.
3. Working on ice, add the following components directly to each well of an optical reaction plate:

Component	Volume for one reaction	Notes
4X TaqMan® Fast Virus 1-Step Master Mix	5 µL	—
TaqMan® Gene Expression Assay (20X)	1 µL	If you are not using pre-formulated TaqMan® Gene Expression Assays, Applied Biosystems recommends primer concentrations of 400 to 900 nM and a probe concentration of 100 to 250 nM.
Sample	Variable	Use as much sample as needed, up to the maximum allowed by the reaction volume.
RT-PCR Grade Water	Variable	Fill to the total reaction volume.
<b>Total volume per reaction</b>	<b>20 µL</b>	—

4. Cover the reaction plate with an optical adhesive cover, vortex the plate briefly, then centrifuge to spin down the contents and eliminate air bubbles.



#### IMPORTANT!

The TaqMan Fast Virus 1-Step Master Mix is a 4X formulation and is more viscous than most master mixes. Be sure that all of the components are thoroughly mixed before proceeding.

## Perform the RT-PCR run

Run the reaction plate using the following run parameters:

### For sample volumes ≤30 µL

Run mode	Default†				
Thermal cycling conditions	Step	Stage	No. of cycles	Temperature	Time
	Reverse transcription	1	1	50 °C‡	5 minutes
	RT inactivation/initial denaturation	2	1	95 °C	20 seconds
	Amplification	3	40	95 °C	3 seconds
60 °C				30 seconds	

† Use the default run mode for your system and sample block module (that is, Fast mode on Fast instruments and standard mode on standard instruments).

‡ Reverse transcription works best between 48 °C and 55 °C.

## Analyze the data

Data analysis varies, depending on the real-time PCR system. For further information, refer to the:

- *TaqMan® Fast Virus 1-Step Master Mix Protocol* (PN 4453800)
- Presence/absence assay (also referred to as *plus/minus assay*) or absolute quantitation assay Getting Started Guide for your real-time PCR system

## Running a no-RT control

If you are concerned that your one-step real-time RT-PCR is detecting genomic DNA rather than a particular RNA species, you can run a no-RT control reaction with the TaqMan Fast Virus 1-Step Master Mix. To run a no-RT control: Heat-kill the RT enzyme by heating one aliquot of master mix at 95 °C for 5 minutes before mixing it with the sample and primer/probe set. The PCR hot-start mechanism will reactivate after the master mix has cooled to room temperature.

## Master mix part numbers and storage conditions

Product	Quantity/part number			Storage conditions
	1 × 1 mL	5 × 1 mL	1 × 10 mL	
TaqMan® Fast Virus 1-Step Master Mix	4444432	4444434	4444436	Store at –20 °C.†

† The TaqMan Fast Virus 1-Step Master Mix will not freeze at –20 °C; gelling may occur.

## Safety information

- !** **IMPORTANT!**
- For safety and biohazard guidelines, refer to the “Safety” section in the *TaqMan® Fast Virus 1-Step Master Mix Protocol* (PN 4453800). For every chemical, read the Safety Data Sheet (SDS) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

## 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](http://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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NOTICE TO PURCHASER: PLEASE REFER TO THE TAQMAN® FAST VIRUS 1-STEP MASTER MIX PROTOCOL (PN 4453800) FOR LIMITED LABEL LICENSE OR DISCLAIMER INFORMATION.

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### Headquarters

850 Lincoln Centre Drive | Foster City, CA 94404 USA  
Phone 650.638.5800 | Toll Free 800.345.5224  
[www.appliedbiosystems.com](http://www.appliedbiosystems.com)

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