

USER GUIDE

applied
biosystems®
by *life* technologies™

TaqMan® Fast Universal PCR Master Mix (2×), No AmpErase® UNG

for use with:

TaqMan® Gene Expression Assays

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About This Guide

IMPORTANT! Before using this product, read and understand the information the “Safety” appendix in this document.

Purpose

The *TaqMan® Fast Universal PCR Master Mix (2X) User Guide* provides reference information for the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG and provides:

- Background information about fast gene quantification assays.
- A list of equipment and materials for using the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG .
- Procedures for using the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG .

For detailed information about specific procedures outlined in this user guide, consult [“Documentation and Support” on page 31](#).

User attention words

Four user attention words may appear in this document. Each word implies a particular level of observation or action as described below:

Note: Provides information that may be of interest or help but is not critical to the use of the product.

IMPORTANT! Provides information that is necessary for proper instrument operation or accurate chemistry kit use.



CAUTION! Indicates a potentially hazardous situation that, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.



WARNING! Indicates a potentially hazardous situation that, if not avoided, could result in death or serious injury.



TaqMan[®] Fast Universal PCR Master Mix (2X), No AmpErase[®] UNG User Guide

Purpose of the product

Use the TaqMan[®] Fast Universal PCR Master Mix (2X), No AmpErase[®] UNG to run TaqMan[®] Gene Expresssays on:

- Applied Biosystems 7900HT Fast Real-Time PCR System with Fast 96-Well Block Module in about 35 minutes.
- Applied Biosystems 7500 Fast Real-Time PCR System in under 40 minutes.
or
- Applied Biosystems 7500 Fast Real-Time PCR System in under 30 minutes, depending on the number of filters selected using Expert Mode.

Kit contents and storage

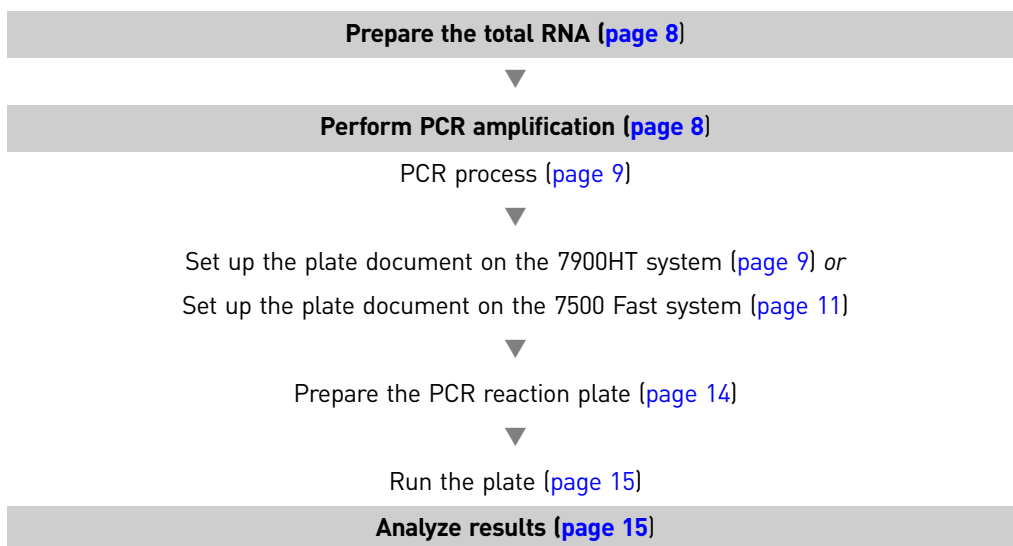
Kit part number	Description	Volume (mL)	No. of 20- μ L reactions	Storage conditions
4352042	TaqMan [®] Fast Universal PCR Master Mix (2X), No AmpErase [®] UNG	2.5 mL	250	2°C to 8°C
4367846		50 mL	5000	

Prepare for RT-PCR

Before you begin the procedure, review the following appendices:

- [Appendix A, “Chemistry Overview” on page 19](#)
- [Appendix B, “Preventing Contamination” on page 23](#)
- [Appendix C, “Materials and Equipment not Included” on page 25](#)

Workflow



Prepare the total RNA

Before using the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG, for fast gene quantification, synthesize single-stranded cDNA from total RNA samples using the High-Capacity cDNA Archive Kit or TaqMan® Reverse Transcription Reagents. See Table 1 on page 25 for ordering information.

Note: Other reverse transcription methods have not been tested for use with the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG.

Perform PCR amplification

During PCR amplification, the hot-start DNA polymerase in the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG:

- Amplifies target cDNA using sequence-specific primers
- Cleaves TaqMan® probes hybridized to the target sequence

The cleavage of TaqMan® probes by the hot-start DNA polymerase generates fluorescent signals detected during PCR.

IMPORTANT! Perform the PCR step described in this protocol on the Applied Biosystems 7500 Fast Real-Time PCR System or the Applied Biosystems 7900HT Fast Real-Time PCR System with Fast 96-Well Block Module.

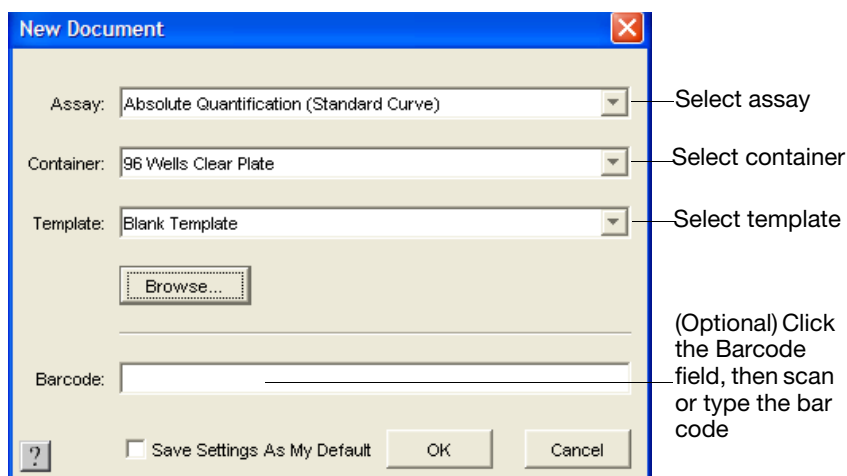
PCR process

1. “Set up the plate document on the 7900HT system” on page 9 or “Set up the plate document on the 7500 Fast system” on page 11
2. “Prepare the PCR reaction plate” on page 14
3. “Run the plate” on page 15

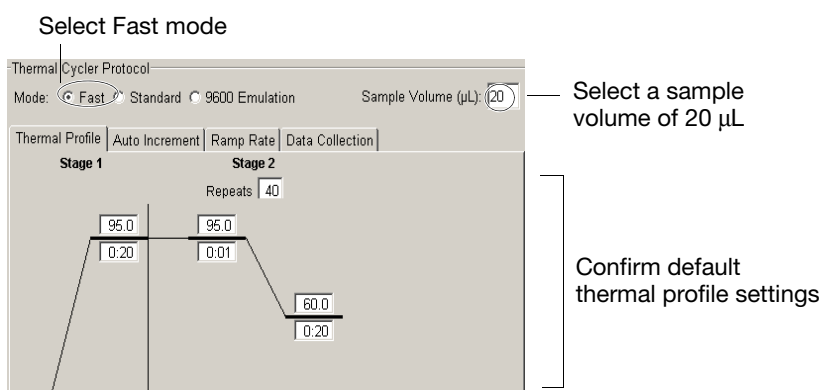
Set up the plate document on the 7900HT system

Note: If using the Applied Biosystems 7900HT Fast Real-Time PCR System, refer to the *Applied Biosystems 7900HT Fast Real-Time PCR System User Bulletin: Performing Fast Gene Quantification* (Part no. 4352533) for detailed instructions on how to create the plate document using the SDS Software v2.2.1 or higher for the 7900HT System.

1. Create a plate document:
 - a. Select **File > New**
 - b. Complete the New Document dialog box, then click **OK**:

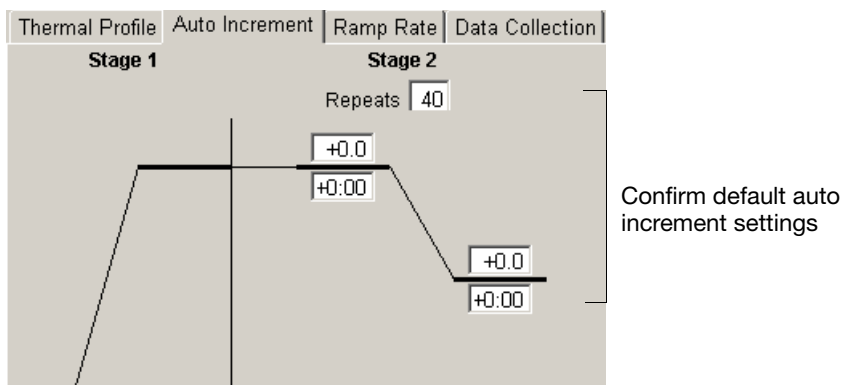


2. Verify that the default settings are used for the thermal cycler protocol:
 - Thermal Profile tab:

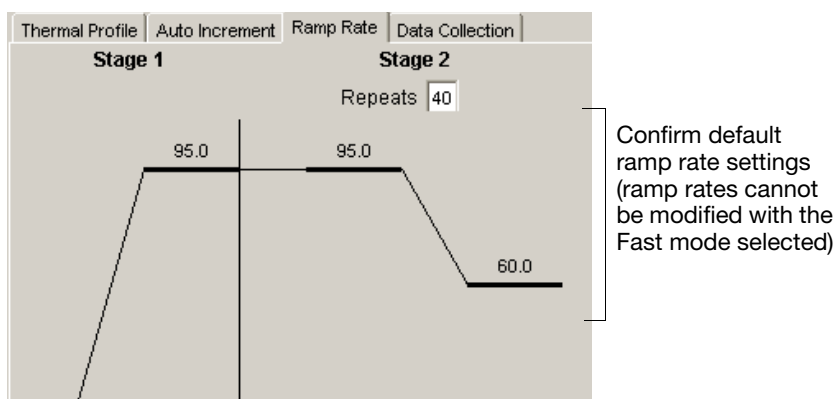


Note: If you use UNG, add an AmpErase® UNG activation step to the beginning of the thermal profile: 50°C for 2 minutes.

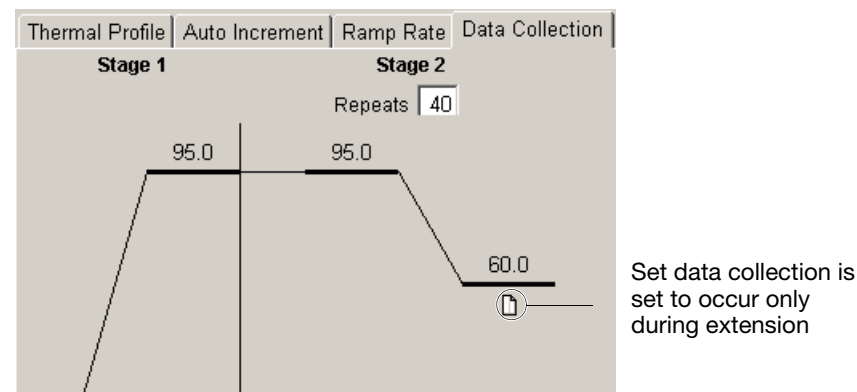
- Auto Increment tab:



- Ramp Rate tab:



- Data Collection tab:



- Complete the setup of the plate document as described in your instrument user guide:
 - Create detectors (the first time only).
 - Copy detectors to the plate document.
 - Apply detectors for samples.
 - Add sample names.
- Save the plate document.

Set up the plate document on the 7500 Fast system

Note: If using the Applied Biosystems 7500 Fast Real-Time PCR System, refer to the *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Absolute Quantification Getting Started Guide* (Part no. 4347825) or the *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Relative Quantification Getting Started Guide* (Part no. 4347824) for detailed instructions on how to create the plate document using the SDS Software v1.3 or higher for the 7500 Fast System.

1. Create a plate document for your application as described in the appropriate 7500 Fast System *Getting Started Guide* listed above:
 - a. Select **File > New**
 - b. Complete the New Document dialog box, then click **Next**:

New Document Wizard

Define Document
Select the assay, container, and template for the document, and enter the operator name and comments.

Assay : Absolute Quantification (Standard Curve) ▼

Container : 96-Well Clear ▼

Template : Blank Document ▼

Run Mode : Fast 7500 ▼

Operator : scientist

Comments : SDS v1.3.1

Plate Name : Plate1

< Back **Next >** Finish Cancel

2. Complete the setup of the plate document for your application as described in the appropriate 7500 Fast System *Getting Started Guide*:
 - a. Create detectors (the first time only).
 - b. Add detectors to the plate document.
 - c. Apply detectors for samples.
 - d. Add sample names.

3. Select the **Instrument** tab, then verify that the default settings are used for the thermal cycler protocol:

- Thermal Profile tab:

Set the default thermal profile settings

Enter sample volume of 20 µL

Select fast mode

Note: If you use UNG, add an AmpErase® UNG activation step to the beginning of the thermal profile: 50°C for 2 minutes.

Note: Optionally, select Expert Mode, which can perform quantification in under 30 minutes, depending on the number of filters selected. For more details, refer to the 7500 Fast System *Getting Started Guide* appropriate for your application.

- Auto Increment tab:

Confirm default auto increment settings

- Ramp Rate tab:

Confirm default ramp rate settings (ramp rates cannot be modified with Fast mode selected)



4. Save the document.

cDNA template guidelines

For optimal performance of TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG, use 10 ng to 100 ng of cDNA per 20-µL reaction.

Reagent preparation guidelines for optimal PCR performance

- Keep all TaqMan® probes protected from light, in the freezer, until you are ready to use them. Excessive exposure to light may affect the fluorescent probes.
- Prior to use:
 - Mix the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG, thoroughly by swirling the bottle.
 - Vortex the TaqMan® Gene Expression Assay, then centrifuge the tube briefly.
 - Place frozen cDNA samples on ice to thaw. After the samples are thawed, vortex them, then centrifuge the tubes briefly.
- Prepare the PCR reaction mix before transferring it to the reaction plate for thermal cycling and fluorescence analysis.

PCR reaction components

For the recommended reaction volume of 20-µL for the Optical 96-Well Fast Plate, each PCR reaction contains the components as listed in the following table.

Component	Volume (µL) / 20-µL reaction
TaqMan® Gene Expression Assay Mix (20X) containing: • Forward PCR primer (18 µM) • Reverse PCR primer (18 µM) • TaqMan® probe (5 µM)	Total reagent volume should be 1.0-µL
cDNA template (10 to 100 ng of cDNA)	10–100 ng [†]
TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG	10.0 µL
RNase-free water	see below [‡]
Total Volume	20.0

[†] If you use UNG, decrease the volume of cDNA template and RNase-free water to 8.8 µL per 20-µL reaction and add 0.2 µL of UNG stock (1 U/µL).

[‡] The volume of RNase-free water (µL) will be 9.0 minus the cDNA sample volume.

Prepare the PCR reaction plate

We recommend performing four replicates of each reaction. For the recommended reaction volume of 20 µL for the Optical 96-Well Fast Plate, prepare the PCR reactions according to the following procedure.

To prepare the reaction plate:

1. Prepare the reaction mix for each sample (for four 20-µL reaction):

Component	Volume (µL) / 20-µL reaction [†]
TaqMan® Gene Expression Assay Mix (20X)	5.0 µL
cDNA template (10 to 100 ng of cDNA) + RNase-free water	45.0 µL [‡]
TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG	50 µL
Total Volume	20.0

[†] Volumes are calculated for five reactions to provide excess volume for the loss that occurs during reagent transfers.

[‡] If you use UNG, add 1.0 µL of UNG stock (1 U/µL) and adjust the amount of cDNA template and RNase-free water.

IMPORTANT! Keep the TaqMan® Gene Expression Assay Mix components protected from direct exposure to light. Excessive exposure to light may affect the fluorescent probes

2. Cap the tube(s), mix by gentle inversion, then centrifuge the tube(s) briefly to spin down the contents and eliminate air bubbles.
3. Transfer 20 µL of reaction mix to the wells of an Optical 96-Well Fast Plate.
Note: The arrangement of the reactions (samples and assays) on the plate must match the arrangement (sample names and detectors/markers) in the plate document used for the run.
4. Seal the plate with an optical adhesive cover, then centrifuge the plate briefly to spin down the contents and eliminate air bubbles.

IMPORTANT! Optical 96-Well Fast Reaction Plates are designed only to be used with optical adhesive covers. Do not use domed or flat caps with Optical 96-Well Fast Reaction Plates.

IMPORTANT! The TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG, does provide a hot-start capability. However, to ensure optimal results, we recommend running the reaction plate as soon as possible after completing the reaction setup. If you cannot run a reaction plate within 2 hours after completing the reaction setup, refrigerate or freeze the reaction plate until you can load and run it on the 7500 Fast System or the 7900HT System.



Run the plate

Note: See the *Applied Biosystems 7900HT Fast Real-Time PCR System User Bulletin: Performing Fast Gene Quantification* (Part no. 4352533), the *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Relative Quantification Getting Started Guide* (Part no. 4347824) or the *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Absolute Quantification Getting Started Guide* (Part no. 4347825) for detailed instructions on loading and running the plate.

1. In the SDS Software for your Real-Time PCR System, open the plate document that corresponds to the reaction plate.
2. Load the reaction plate into the 7500 Fast System or the 7900HT System.
3. Start the run.

Analyze results

General process

The general process for analyzing the data from gene expression assays involves the following procedures:

1. View the amplification plots.
2. Set the baseline and threshold values.
3. Use the standard curve method or the relative quantification ($\Delta\Delta C_T$) method to analyze the results.

Resources for data analysis

Refer to the following documents for more information about analyzing your data:

- *Applied Biosystems 7900HT Fast Real-Time PCR System and SDS Enterprise Database User Guide* (Part no. 4351684)
- *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Relative Quantification Getting Started Guide* (Part no. 4347824)
- *Applied Biosystems 7300/7500/7500 Fast Real-Time PCR Systems Absolute Quantification Getting Started Guide* (Part no. 4347825)
- *Real-Time PCR Systems Chemistry Guide: Applied Biosystems 7900HT Fast Real-Time PCR Systems and 7300/7500/7500 Fast Real-Time PCR Systems* (Part no. 4348358)

Troubleshooting

Observation	Possible Cause	Action
High C_T values, poor precision, or failed PCR reactions	Insufficient cDNA template is present	Use 10 to 100 ng of cDNA template per 20- μ L reaction.
	Quality of cDNA template is poor	1. Quantify the amount of cDNA template. 2. Test the cDNA template for the presence of PCR inhibitors.
	Sample degradation	Prepare fresh cDNA, then repeat the experiment.
	The TaqMan® Universal PCR Master Mix (2X) was used instead of the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG	Prepare the reactions as described on page 13 .
	Reduced number of PCR cycles in the thermal cycler protocol	Increase the number of PCR cycles to the default setting of 40 (see page 9).
Low ΔR_n or R_n values	Primer-dimer formation and residual polymerase activity	1. Prepare the reaction mixes and the reaction plate on ice. 2. Run the reaction plate as soon as possible after completing the reaction setup. If you cannot run a reaction plate within 2 hours after completing the reaction setup, refrigerate or freeze the reaction plate until you can load and run it on the 7900HT or 7500 Fast instruments.
	Extension time is too short	Use the default thermal profile settings (see page 9).
Run takes more than 40 minutes	Thermal cycler mode is set to Standard or 9600 Emulation	Make sure that the thermal cycler mode is set to Fast (see page 9).
R_n vs. Cycle plot is not displayed	ROX™ dye was not selected as the passive reference when the plate document was set up	Select ROX dye as the passive reference when you set up the plate document.

Observation	Possible Cause	Action
Extremely high ΔR_n or R_n values	ROX dye was not selected as the passive reference when the plate document was set up	Select ROX dye as the passive reference when you set up the plate document.
	Evaporation	Make sure that the reaction plate is sealed completely, especially around the edges.
Lower ΔR_n values obtained in early cycles	C_T value is less than 15	Adjust the upper baseline range to a value less than 15.
High variability across the reaction plate	ROX dye was not selected as the passive reference when the plate document was set up	Select ROX dye as the passive reference when you set up the plate document.
	Evaporation	Make sure that the reaction plate is sealed completely, especially around the edges.
High variability across replicates	Reaction mix was not mixed well	Mix the reaction mix gently by inversion, then centrifuge briefly before aliquoting to the reaction plate.
Poor amplification of target in multiplex assay when using: - TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG - Default Fast thermal cycling conditions	Target is difficult to amplify	Increase the annealing/extension temperature to 62°C. Increase the annealing/extension time in the thermal cycler protocol to 30 seconds. If you do not obtain acceptable performance by increasing both the annealing/extension temperature and time, you may need to reoptimize assay components. Refer to the <i>Real-Time PCR Systems Chemistry Guide</i> (Part no. 4348358) for conditions under which you should optimize.





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Chemistry overview

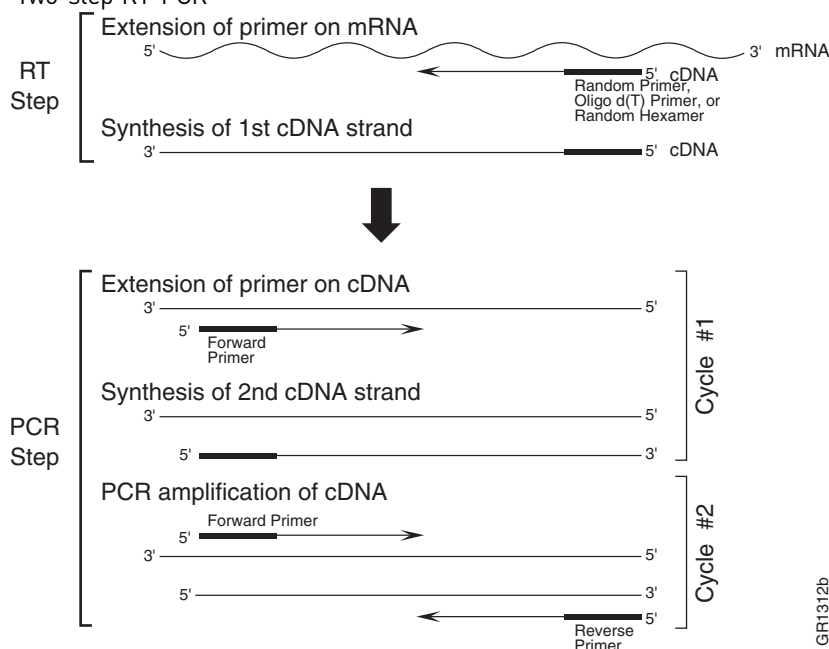
Two-step PCR

Gene quantification assays using TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG, and TaqMan® Gene Expression Assays are performed in a two-step RT-PCR:

1. In the reverse transcription (RT) step, cDNA is reverse transcribed from total RNA samples using random primers from the High-Capacity cDNA Archive Kit or using oligo d(T) primers or random hexamers from the TaqMan® Reverse Transcription Reagents.
2. In the PCR step, PCR products are synthesized from cDNA samples using the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG.

Note: Figure 1 does not show hybridization of the TaqMan MGB probe (labeled with FAM™ dye).

Figure 1 Two-step RT-PCR



GR1312b

About DNA polymerase

The TaqMan[®] Fast Universal PCR Master Mix (2X), No AmpErase[®] UNG, contains a hot-start DNA polymerase system that does not require an activation step. Performance is similar to that of the AmpliTaq Gold[®] DNA Polymerase.

About the probes

The TaqMan[®] MGB probes contain:

- A reporter dye (for example, FAM dye) linked to the 5' end of the probe.
- A minor groove binder (MGB) at the 3' end of the probe. MGBs increase the melting temperature (T_m) without increasing probe length (Afonina *et al.*, 1997; Kutyavin *et al.*, 1997); they also allow the design of shorter probes.
- A nonfluorescent quencher (NFQ) at the 3' end of the probe. Because the quencher does not fluoresce, Applied Biosystems Real-Time PCR Systems can measure reporter dye contributions more accurately.

5' Nuclease assay process

The 5' nuclease assay process (Figures 2 through 6) takes place during PCR amplification. This process occurs in every cycle and does not interfere with the exponential accumulation of product.

Figure 2 Legend for 5' nuclease assay process figures

-  = Nonfluorescent quencher
-  = Minor groove binder
-  = Reporter
-  = Hot-start DNA polymerase system

During PCR, the TaqMan[®] MGB probe anneals specifically to a complementary sequence between the forward and reverse primer sites (Figure 3).

When the probe is intact (Figures 3 and 4), the proximity of the reporter dye to the quencher dye results in suppression of the reporter fluorescence primarily by Förster-type energy transfer (Förster, 1948; Lakowicz, 1983).

Figure 3 Polymerization

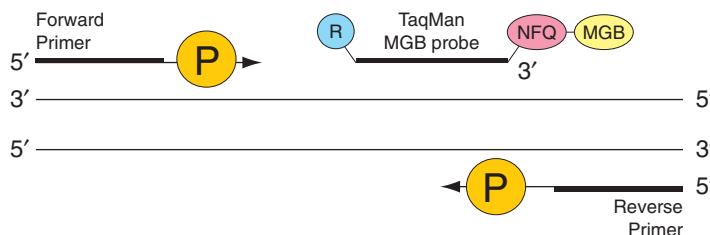
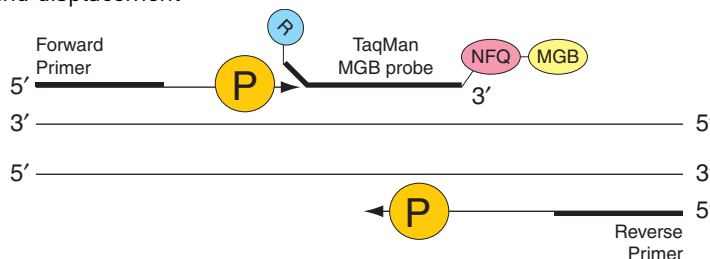
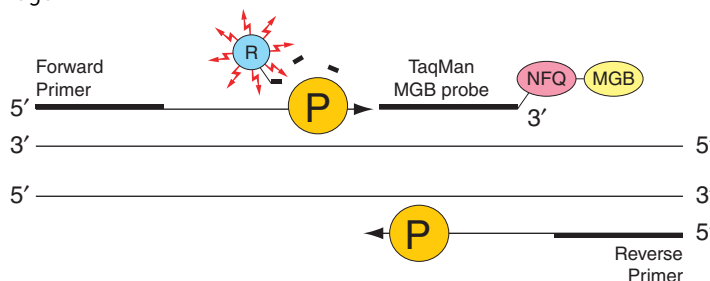


Figure 4 Strand displacement



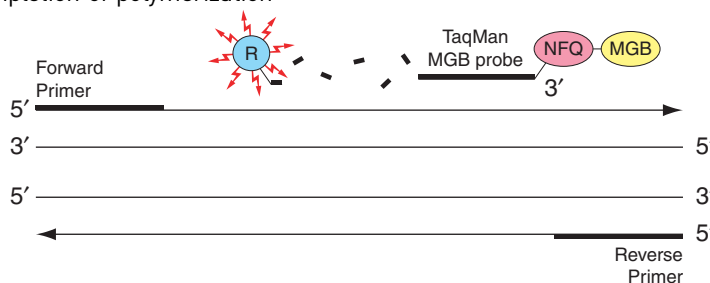
The DNA polymerase cleaves only probes that are hybridized to the target (Figure 5). Cleavage separates the reporter dye from the quencher dye; resulting in increased fluorescence by the reporter. The increase in fluorescence signal occurs only if the target sequence is complementary to the probe and is amplified during PCR. Because of these requirements, nonspecific amplification is not detected.

Figure 5 Cleavage



Polymerization of the strand continues, but because the 3' end of the probe is blocked, there is no extension of the probe during PCR (Figure 6).

Figure 6 Completion of polymerization





Appendix A Chemistry Overview

Chemistry overview

Overview

PCR assays require special laboratory practices to avoid false positive amplifications (Kwok and Higuchi, 1989). The high throughput and repetition of these assays can lead to amplification of a single DNA molecule (Saiki *et al.*, 1985; Mullis and Faloona, 1987).

General PCR practices

General PCR practices to prevent contamination:

- Maintain separate areas, dedicated equipment, and supplies for:
 - Sample preparation
 - PCR setup
 - PCR amplification
 - Analysis of PCR products
- Do not bring amplified PCR products into the PCR setup area.
- Wear a clean lab coat (not previously worn while handling amplified PCR products or used during sample preparation) and clean gloves when preparing samples for PCR amplification.
- Change gloves whenever you suspect that they are contaminated.
- Open and close all sample tubes and reaction plates carefully. Try not to splash or spray PCR samples.
- Keep reactions and components capped as much as possible.
- Use positive-displacement or aerosol-resistant pipette tips.
- Clean lab benches and equipment periodically with freshly diluted 10% bleach solution.



Materials and Equipment not Included

The following table includes required and optional equipment and materials for using the TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG . Unless otherwise noted, many items listed are available from major laboratory suppliers (MLS).

Materials and Equipment	Source
Applied Biosystems 7900HT Fast Real-Time PCR System with Fast 96-Well Block Module	Contact your Life Technologies sales representative.
7900HT System Fast Service Upgrade	
Applied Biosystems 7500 Fast Real-Time PCR System	
7500 Fast Real-Time PCR Upgrade Kit	
SDS Software v2.2.1 or higher for the 7900HT System	Contact your Life Technologies sales representative.
SDS Software v1.3 or higher for the 7500 Fast System	
Microsoft® Excel® or equivalent spreadsheet and analysis software	Software suppliers
TaqMan® Fast Reagents Starter Kit	Life Technologies (Part no. 4352407)
TaqMan® Fast Universal PCR Master Mix (2X), No AmpErase® UNG	Life Technologies (Part no. 4352042)
AmpErase® Uracil N-glycosylase (UNG)	Life Technologies (Part no. N8080096)
TaqMan® Gene Expression Assays	Life Technologies (Part no. 4331182 and Part no. 4351372)
TaqMan® Reverse Transcription Reagents	Life Technologies (Part no. N8080234)
High-Capacity cDNA Archive Kit	Life Technologies (Part no. 4322171)
RNase-free, sterile-filtered water	MLS
ABI Prism® Optical Adhesive Cover Starter Kit	Life Technologies (Part no. 4313663)
ABI Prism® Optical Adhesive Covers (quantity 100)	Life Technologies (Part no. 4311971)
ABI Prism® Optical Adhesive Covers (quantity 25)	Life Technologies (Part no. 4360954)
Optical 96-Well Fast Thermal Cycling Plates with Barcode (code 128), 20 plates	Life Technologies (Part no. 4346906)

Materials and Equipment	Source
Reagent Tubes With Caps, 10-mL	Life Technologies (Part no. 4305932)
Accessories for opening tubes of TaqMan® Gene Expression Assays: Decapper for single caps (Part no. 54000) Decapper for eight caps (Part no. 54001) TPE cap cluster for simultaneously capping of 96 individual polypropylene tubes, 50 capmats/bag (Part no. 53001)	Micronic BV† PO Box 604 8200 AP Lelystad Netherlands Telephone: +31(0)320.277077 Fax: +31(1)320.277088
Centrifuge with plate holders	MLS
Disposable gloves	MLS
Microcentrifuge	MLS
Pipette tips, aerosol resistant, nuclease-free: 1- to 20- μ L range, 20- to 200- μ L range, 100- to 1000- μ L range	MLS
Pipettors (positive-displacement, air-displacement, or multichannel): 1- to 20- μ L range, 20- to 200- μ L range, 100- to 1000- μ L range	MLS
Polypropylene tubes	MLS
Vortexer	MLS

† Other vendors supply similar products.



WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
 - Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, etc). To obtain SDSs, see the “Documentation and Support” section in this document.
-

Chemical safety



WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below, and consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the “Documentation and Support” section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer's cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if necessary) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.



WARNING! 4L Reagent and Waste Bottle Safety. Four-liter reagent and waste bottles can crack and leak. Each 4-liter bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position.

Biological hazard safety



WARNING! Potential Biohazard. Depending on the samples used on this instrument, the surface may be considered a biohazard. Use appropriate decontamination methods when working with biohazards.



WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Follow all applicable local, state/provincial, and/or national regulations. Wear appropriate protective equipment, which includes but is not limited to: protective eyewear, face shield, clothing/lab coat, and gloves. All work should be conducted in properly equipped facilities using the appropriate safety equipment (for example, physical containment devices). Individuals should be trained according to applicable regulatory and company/institution requirements before working with potentially infectious materials. Read and follow the applicable guidelines and/or regulatory requirements in the following:

In the U.S.:

- U.S. Department of Health and Human Services guidelines published in Biosafety in Microbiological and Biomedical Laboratories found at: www.cdc.gov/biosafety
- Occupational Safety and Health Standards, Bloodborne Pathogens (29 CFR§1910.1030), found at: www.access.gpo.gov/nara/cfr/waisidx_01/29cfr1910a_01.html
- Your company's/institution's Biosafety Program protocols for working with/handling potentially infectious materials.
- Additional information about biohazard guidelines is available at: www.cdc.gov

In the EU:

Check local guidelines and legislation on biohazard and biosafety precaution and refer to the best practices published in the World Health Organization (WHO) Laboratory Biosafety Manual, third edition, found at: www.who.int/csr/resources/publications/biosafety/WHO_CDS_CSR_LYO_2004_11/en/



Appendix D Safety
Biological hazard safety

Documentation and Support

Related documentation

For detailed information about specific procedures outlined in this user guide, consult the following documents:

Document	Part number
<i>Applied Biosystems 7900HT Fast Real-Time PCR System and SDS Enterprise Database User Guide</i>	4351684
<i>Applied Biosystems 7900HT Fast Real-Time PCR System Quick Reference: Performing Fast Gene Quantification</i>	4351892
<i>Applied Biosystems 7900HT Fast Real-Time PCR System User Bulletin: Performing Fast Gene Quantification</i>	4352533
<i>Applied Biosystems 7300/7500/7500 Fast Real-Time PCR System Installation and Maintenance Getting Started Guide</i>	4347828
<i>Applied Biosystems 7300/7500/7500 Fast Real-Time PCR System Absolute Quantification Getting Started Guide</i>	4347825
<i>Applied Biosystems 7300/7500/7500 Fast Real-Time PCR System Relative Quantification Getting Started Guide</i>	4347824
<i>Applied Biosystems 7500 Fast Real-Time PCR System Quick Reference: Performing Fast Gene Quantification</i>	4362285
<i>Real-Time PCR Systems Chemistry Guide</i>	4348358
<i>TaqMan® Gene Expression Assays User Guide</i>	4333458
<i>Applied Biosystems 7900HT Fast Real-Time PCR System and SDS Enterprise Database User Guide</i>	4351684

Note: For additional documentation, see [“Obtaining support” on page 32](#).

A fast gene quantification procedural overview is also provided in a Quick Reference: *Performing Fast Gene Quantification for the 7900HT System Quick Reference* (Part no. 4351892) and the 7500 Fast System (Part no. 4362285).

Obtaining SDSs

Safety Data Sheets (SDSs) are available from www.appliedbiosystems.com/sds

Note: For the SDSs of chemicals not distributed by Life Technologies, contact the chemical manufacturer.

Obtaining support

For the latest services and support information for all locations, go to:

www.lifetechnologies.com

At the website, you can:

- Access worldwide telephone and fax numbers to contact Technical Support and Sales facilities
- Search through frequently asked questions (FAQs)
- Submit a question directly to Technical Support
- Search for user documents, SDSs, vector maps and sequences, application notes, formulations, handbooks, certificates of analysis, citations, and other product support documents
- Obtain information about customer training
- Download software updates and patches

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