January 2014

Rotor-Gene® Probe Handbook

Rotor-Gene Probe PCR Kit

Rotor-Gene Probe RT-PCR Kit

For fast real-time PCR, two-step RT-PCR, and one-step RT-PCR using sequence-specific probes on Rotor-Gene cyclers

Sample & Assay Technologies

QIAGEN Sample and Assay Technologies

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Contents

Kit Contents

Rotor-Gene Probe PCR Kit	**(400)**
Catalog no.	**204374**
Number of 25 µl reactions	**400**
2x Rotor-Gene Probe PCR Master Mix, containing: ■ HotStarTaq® *Plus* DNA Polymerase ■ Rotor-Gene Probe PCR Buffer ■ dNTP mix (dATP, dCTP, dGTP, dTTP)	3 x 1.7 ml
RNase-Free Water	2 x 2 ml
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Rotor-Gene Probe RT-PCR Kit	**(400)**
Catalog no.	**204574**
Number of 25 µl reactions	**400**
2x Rotor-Gene Probe RT-PCR Master Mix, containing: ■ HotStarTaq *Plus* DNA Polymerase ■ Rotor-Gene Probe RT-PCR Buffer ■ dNTP mix (dATP, dCTP, dGTP, dTTP)	3 x 1.7 ml
Rotor-Gene RT Mix, a mixture of the QIAGEN® products: ■ Omniscript® Reverse Transcriptase ■ Sensiscript® Reverse Transcriptase	100 µl
RNase-Free Water	2 x 2 ml
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Shipping and Storage

Rotor-Gene Probe Kits are shipped on dry ice. The kits should be stored immediately upon receipt at –15°C to –30°C and protected from light. When the kits are stored under these conditions and handled correctly, performance is guaranteed until the expiration date (see the quality-control label inside the kit box or on the kit envelope). 2x Rotor-Gene Probe Master Mixes can also be stored protected from light at 2–8°C for up to 2 months without showing any reduction in performance.

Product Use Limitations

Rotor-Gene Probe Kits are intended for molecular biology applications. These products are not intended for the diagnosis, prevention, or treatment of a disease.

All due care and attention should be exercised in the handling of the products. We recommend all users of QIAGEN products to adhere to the NIH guidelines that have been developed for recombinant DNA experiments, or to other applicable guidelines.

Product Warranty and Satisfaction Guarantee

QIAGEN guarantees the performance of all products in the manner described in our product literature. The purchaser must determine the suitability of the product for its particular use. Should any product fail to perform satisfactorily due to any reason other than misuse, QIAGEN will replace it free of charge or refund the purchase price. We reserve the right to change, alter, or modify any product to enhance its performance and design. If a QIAGEN product does not meet your expectations, simply call your local Technical Service Department or distributor. We will credit your account or exchange the product — as you wish. Separate conditions apply to QIAGEN scientific instruments, service products, and to products shipped on dry ice. Please inquire for more information.

A copy of QIAGEN terms and conditions can be obtained on request, and is also provided on the back of our invoices. If you have questions about product specifications or performance, please call QIAGEN Technical Services or your local distributor (see back cover or visit www.qiagen.com).

Technical Assistance

At QIAGEN, we pride ourselves on the quality and availability of our technical support. Our Technical Service Departments are staffed by experienced scientists with extensive practical and theoretical expertise in sample and assay technologies and the use of QIAGEN products. If you have any questions or experience any difficulties regarding Rotor-Gene Probe Kits or QIAGEN products in general, please do not hesitate to contact us.

QIAGEN customers are a major source of information regarding advanced or specialized uses of our products. This information is helpful to other scientists as well as to the researchers at QIAGEN. We therefore encourage you to contact us if you have any suggestions about product performance or new applications and techniques.

For technical assistance and more information, please see our Technical Support Center at www.qiagen.com/Support or call one of the QIAGEN

Technical Service Departments or local distributors (see back cover or visit www.qiagen.com).

Safety Information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). These are available online in convenient and compact PDF format at www.qiagen.com/safety where you can find, view, and print the SDS for each QIAGEN kit and kit component.

24-hour emergency information

Chemical emergency or accident assistance is available 24 hours a day from:

CHEMTREC

USA & Canada ▪ Tel: 1-800-424-9300

Outside USA & Canada ▪ Tel: +1-703-527-3887 (collect calls accepted)

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each component of Rotor-Gene Probe PCR Kits and Rotor-Gene Probe RT-PCR Kits is tested against predetermined specifications to ensure consistent product quality. See the quality-control label inside the kit box or on the kit envelope for lot-specific values.

Product Description

Component	Description
HotStarTaq *Plus* DNA Polymerase*	HotStarTaq *Plus* DNA Polymerase is a modified form of a recombinant 94 kDa DNA polymerase, originally isolated from *Thermus aquaticus*. HotStarTaq *Plus* DNA Polymerase is provided in an inactive state and has no enzymatic activity at ambient temperature. The enzyme is activated by a 3- or 5-minute, 95°C incubation step.
Rotor-Gene Probe PCR or RT-PCR Buffer*	Contains Tris·Cl, KCl, $(NH_4)_2SO_4$, $MgCl_2$; also contains Q-Bond® and other additives that enable fast cycling on Rotor-Gene cyclers
dNTP mix*	Contains dATP, dCTP, dGTP, and dTTP of ultrapure quality
RNase-free water	Ultrapure quality, PCR-grade
Rotor-Gene RT Mix†	Contains an optimized mixture of the QIAGEN products Omniscript Reverse Transcriptase and Sensiscript Reverse Transcriptase, which are recombinant heterodimeric enzymes expressed in *E. coli*.

* Included in 2x Rotor-Gene Probe PCR Master Mix and 2x Rotor-Gene Probe RT-PCR Master Mix.

† Supplied with the Rotor-Gene Probe RT-PCR Kit.

Introduction

Rotor-Gene Probe PCR Kits provide rapid real-time quantification of genomic DNA, cDNA, or RNA targets on the Rotor-Gene Q in an easy-to-handle format. The kits are also compatible with the Rotor-Gene 3000 and Rotor-Gene 6000. Two kit formats are available:

- **Rotor-Gene Probe PCR Kit**: The kit can be used in real-time PCR of genomic DNA targets, and also in real-time two-step RT-PCR of RNA targets following reverse transcription with, for example, the QuantiTect® Reverse Transcription Kit (see ordering information, page 26).
- **Rotor-Gene Probe RT-PCR Kit**: The kit can be used in real-time one-step RT-PCR of RNA targets, with reverse transcription and PCR taking place sequentially in the same tube.

The kits are compatible with TaqMan® probes. High specificity and sensitivity in PCR and RT-PCR are achieved by the use of the hot-start enzyme HotStarTaq *Plus* DNA Polymerase together with a specialized fast PCR or RT-PCR buffer. For one-step RT-PCR, the optimized Omniscript and Sensiscript blend for the reverse-transcription step further enhances sensitivity. Short cycling steps without loss of PCR sensitivity and efficiency are enabled by Q-Bond, a patent-pending additive in the PCR and RT-PCR buffers.

The kits have been optimized for use with Rotor-Gene real-time PCR cyclers, which employ a unique centrifugal rotary design. PCR tubes are placed into a rotor which spins tubes past the same excitation light source and the same detector in a chamber of moving air. This means that there is minimal optical and temperature variation between tubes, enabling high precision in real-time PCR quantification. In addition, as the rotor spins continuously at 400 rpm, high-speed data acquisition is possible.

2x Rotor-Gene Probe Master Mixes

The components of 2x Rotor-Gene Probe PCR Master Mix include HotStarTaq *Plus* DNA Polymerase and Rotor-Gene Probe PCR Buffer (see descriptions below).

2x Rotor-Gene Probe RT-PCR Master Mix contains HotStarTaq *Plus* DNA Polymerase and Rotor-Gene Probe RT-PCR Buffer (see descriptions below).

HotStarTaq *Plus* DNA Polymerase

HotStarTaq *Plus* DNA Polymerase is a modified form of QIAGEN *Taq* DNA Polymerase. It is provided in an inactive state and has no enzymatic activity at ambient temperature.

In real-time PCR and two-step RT-PCR, this inactivity of HotStarTaq *Plus* DNA Polymerase prevents the formation of misprimed products and primer–dimers

during reaction setup and the first denaturation step, leading to high PCR specificity and accurate quantification. The enzyme is activated at the start of a reaction by a 3-minute, 95°C incubation step. The hot start enables reactions to be set up rapidly and conveniently at room temperature.

In real-time one-step RT-PCR, HotStarTaq *Plus* DNA Polymerase remains completely inactive during the reverse-transcription reaction and does not interfere with it. This prevents formation of misprimed RT-PCR products and primer–dimers during reaction setup, reverse transcription, and the first denaturation step. The enzyme is activated after the reverse-transcription step by a 5-minute, 95°C incubation step. The hot start also inactivates the reverse transcriptases, ensuring temporal separation of reverse transcription and PCR, and allowing both steps to be performed sequentially in a single tube.

For all reactions, the concentration of HotStarTaq *Plus* DNA Polymerase in the master mixes is optimized to allow short extension times in the combined annealing/extension step of each PCR cycle.

Rotor-Gene Probe Buffers

Rotor-Gene Probe Buffers are specially optimized to support the Rotor-Gene cyclers' fast-cycling capabilities, which are based on the cyclers' unique centrifugal rotary design. Rotor-Gene Probe PCR Buffer is specifically designed for fast-cycling, real-time PCR using sequence-specific probes. Rotor-Gene Probe RT-PCR Buffer is specifically designed for fast-cycling, real-time one-step RT-PCR using sequence-specific probes. A novel additive in the buffers, Q-Bond, allows short cycling times on Rotor-Gene cyclers. Q-Bond increases the affinity of *Taq* DNA polymerases for short single-stranded DNA, reducing the time required for primer/probe annealing to a few seconds. This allows a combined annealing/extension step of only 10 seconds. In addition, the unique composition of the buffers supports the melting behavior of DNA, enabling short denaturation and annealing/extension times.

Rotor-Gene Probe PCR Buffer is based on the unique QIAGEN PCR buffer system. Rotor-Gene Probe RT-PCR Buffer is based on the unique QIAGEN OneStep RT-PCR buffer system. The buffers contain a balanced combination of KCl and $(NH_4)_2SO_4$, which promotes a high ratio of specific to nonspecific primer binding during the annealing step of each PCR cycle. This creates stringent primer annealing conditions, leading to increased PCR specificity. When using these buffers, primer annealing is only marginally influenced by the $MgCl_2$ concentration, so optimization by titration of Mg^{2+} is not required.

cDNA synthesis for real-time one-step RT-PCR

Use of 2x Rotor-Gene Probe RT-PCR Master Mix together with Rotor-Gene RT Mix allows both reverse transcription and PCR to take place in a single tube. All reagents required for both reactions are added at the beginning, so there is no need to open the tube once the reverse-transcription reaction has been started.

Rotor-Gene RT Mix contains an optimized Omniscript and Sensiscript blend. Both enzymes exhibit a high affinity for RNA, facilitating transcription through secondary structures that may inhibit other reverse transcriptases. Omniscript is designed for reverse transcription of RNA amounts greater than 50 ng, and Sensiscript is optimized for use with very small amounts of RNA (<50 ng). This enzyme combination provides highly efficient and sensitive reverse transcription over a wide range of RNA template amounts.

cDNA synthesis for real-time two-step RT-PCR

If quantifying cDNA targets with the Rotor-Gene Probe PCR Kit, RNA must first be reverse transcribed into cDNA. A portion of the reverse-transcription reaction is then transferred to another tube where real-time PCR takes place. This entire process is known as real-time two-step RT-PCR, since reverse transcription and real-time PCR are carried out in separate tubes.

For reverse transcription, we recommend using the QuantiTect Reverse Transcription Kit. The kit provides a fast and convenient procedure, requiring only 20 minutes to synthesize first-strand cDNA and eliminate genomic DNA contamination. An optimized mix of oligo-dT and random primers enables cDNA synthesis from all regions of RNA transcripts, even from 5' regions of very long mRNA transcripts. cDNA yields are high, allowing sensitive detection of even low-abundance transcripts in real-time two-step RT-PCR. An alternative to the QuantiTect Reverse Transcription Kit is the FastLane® Cell cDNA Kit, which allows cDNA to be prepared directly from cultured cells without RNA purification. The FastLane Cell cDNA Kit is useful for experiments where archiving of purified RNA is not required. For ordering information for these 2 kits, see page 26.

Using the correct protocol

This handbook contains 2 protocols:

- Real-time PCR and two-step RT-PCR using TaqMan probes (page 11)
- Real-time one-step RT-PCR using TaqMan probes (page 16)

For background information on real-time PCR, please refer to "Guidelines for real-time PCR and RT-PCR" at www.qiagen.com/resources/info, which contains guidelines on template preparation, primer design, controls, data analysis, and other topics.

Protocol: Real-Time PCR and Two-Step RT-PCR Using TaqMan Probes

This protocol can be used with the Rotor-Gene Q, Rotor-Gene 3000, or Rotor-Gene 6000.

Important points before starting

- Always start with the cycling conditions specified in this protocol, even if using previously established primer–probe systems.
- For the highest efficiency in real-time PCR using sequence-specific probes, targets should ideally be 70–200 bp in length and should not exceed 300 bp.
- The PCR must start with an **initial incubation step of 3 minutes at 95°C** to activate HotStarTaq *Plus* DNA Polymerase.
- We recommend a final reaction volume of 25 µl.
- **Always start with the Mg^{2+} concentration as provided** in 2x Rotor-Gene Probe PCR Master Mix.

Procedure

1. **Thaw 2x Rotor-Gene Probe PCR Master Mix, template DNA or cDNA, primer and probe solutions, and RNase-free water. Mix the individual solutions.**
2. **Prepare a reaction mix according to Table 1 (page 12).**

   Due to the hot start, it is not necessary to keep samples on ice during reaction setup or while programming the Rotor-Gene cycler.

   Note: We strongly recommend starting with the Mg^{2+} concentration as provided in 2x Rotor-Gene Probe PCR Master Mix.

Table 1. Reaction setup

Component	Volume/reaction	Final concentration
2x Rotor-Gene Probe PCR Master Mix	12.5 µl	1x
Primer A	Variable	0.4 µM
Primer B	Variable	0.4 µM
Probe	Variable	0.2 µM
Template DNA or cDNA (added at step 4)	Variable	≤100 ng/ reaction
RNase-free water	Variable	
Total reaction volume	**25 µl**	

3. **Mix the reaction mix thoroughly, and dispense appropriate volumes into PCR tubes.**
4. **Add template DNA or cDNA (≤100 ng/reaction) to the individual PCR tubes containing the reaction mix.**

   For two-step RT-PCR, the volume of the cDNA added (from the undiluted RT reaction) should not exceed 10% of the final PCR volume.
5. **Program the Rotor-Gene cycler according to the program outlined in Table 2 (page 13) and Figures 1 and 2 (page 14 and 15).**

   Data acquisition should be performed during the combined annealing/ extension step.

Table 2. Cycling conditions

Step	Time	Temperature	Additional comments
PCR initial activation step	3 min	95°C	HotStarTaq *Plus* DNA Polymerase is activated by this heating step
Two-step cycling			
Denaturation	3 s	95°C	
Combined annealing/ extension	10 s	60°C	Perform fluorescence data collection
Number of cycles	35–40		The number of cycles depends on the amount of template DNA

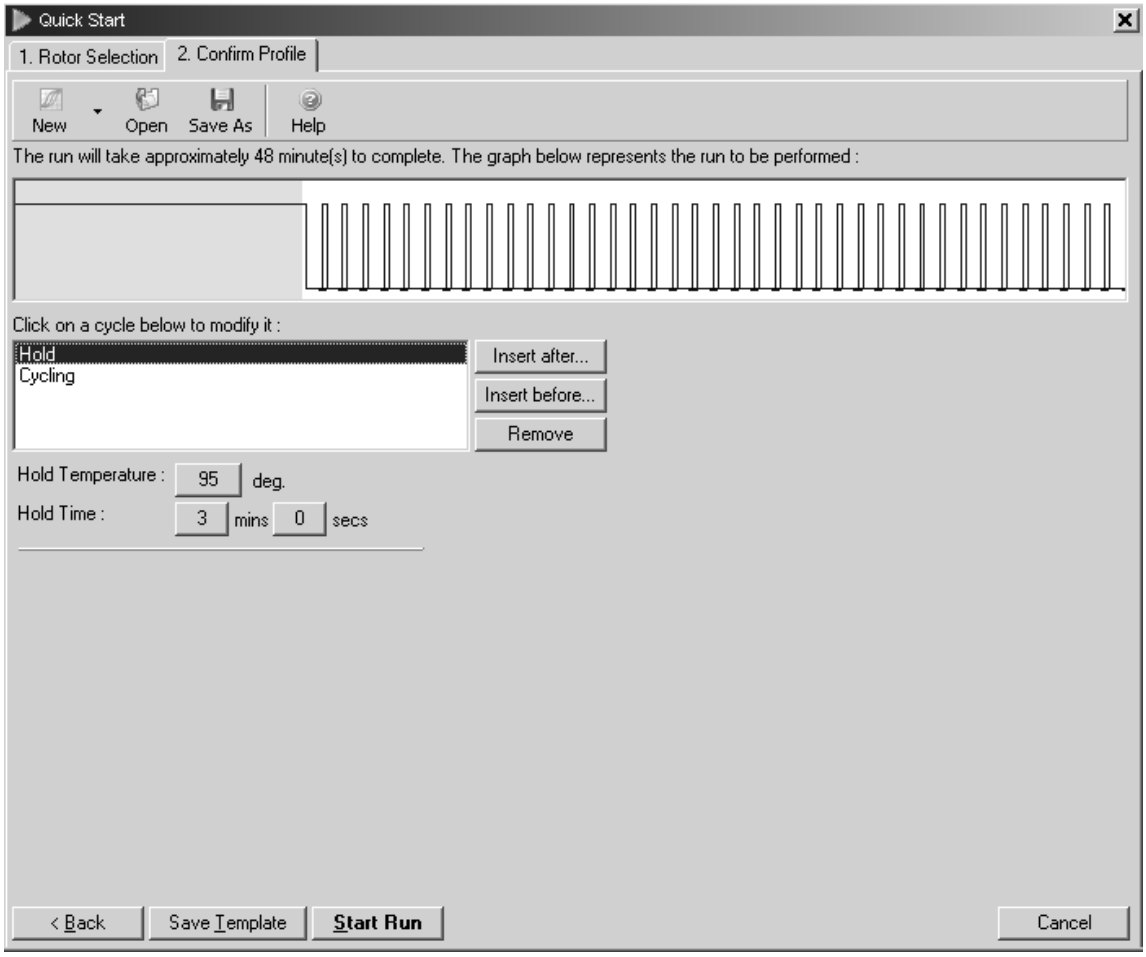

Figure 1. PCR initial activation step. PCR requires an initial incubation at 95°C for 3 min to activate HotStarTaq *Plus* DNA Polymerase.

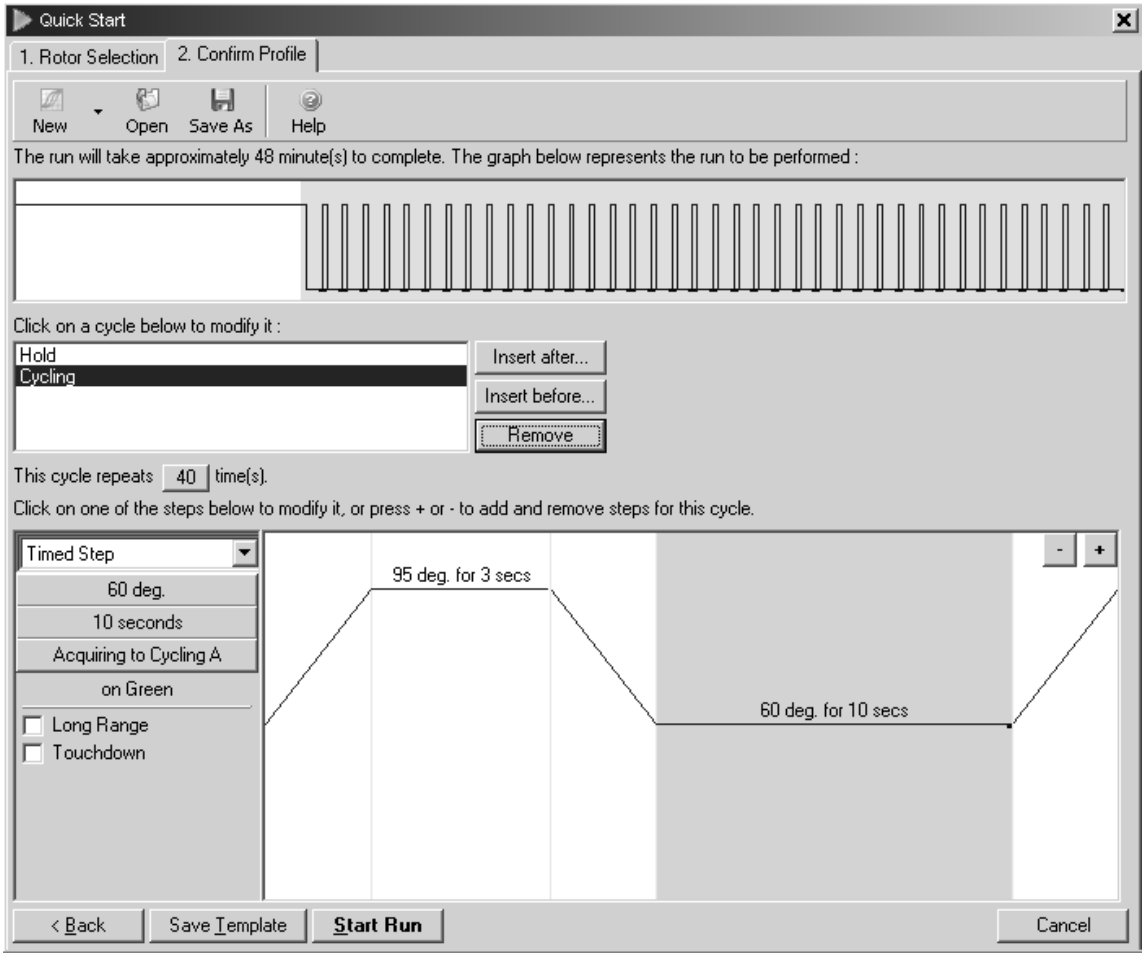

Figure 2. Two-step cycling. PCR requires 35–40 cycles. Each cycle is comprised of 2 steps: 95°C for 3 s (denaturation step) and 60°C for 10 s (annealing/extension step).

6. Place the PCR tubes in the Rotor-Gene cycler, and start the cycling program.

Protocol: Real-Time One-Step RT-PCR Using TaqMan Probes

This protocol can be used with the Rotor-Gene Q, Rotor-Gene 3000, or Rotor-Gene 6000.

Important points before starting

- Always start with the cycling conditions specified in this protocol, even if using previously established primer–probe systems.
- For the highest efficiency in real-time RT-PCR using sequence-specific probes, targets should ideally be 70–200 bp in length and should not exceed 300 bp.
- After reverse transcription, the PCR step of the RT-PCR must start with an **initial incubation step of 5 minutes at 95°C** to activate HotStarTaq *Plus* DNA Polymerase.
- Set up all reactions on ice to avoid premature cDNA synthesis.
- We recommend a final reaction volume of 25 µl.
- **Always start with the Mg^{2+} concentration as provided** in 2x Rotor-Gene Probe RT-PCR Master Mix.

Procedure

1. **Thaw 2x Rotor-Gene Probe RT-PCR Master Mix, template RNA, primer and probe solutions, and RNase-free water. Mix the individual solutions, and place them on ice. Rotor-Gene RT Mix should be taken from –15°C to –30°C immediately before use, always kept on ice, and returned to storage at –15°C to –30°C immediately after use.**
2. **Prepare a reaction mix according to Table 3 (page 17).**

   Keep samples on ice while preparing the reaction mix.

   Note: We strongly recommend starting with the Mg^{2+} concentration as provided in 2x Rotor-Gene Probe RT-PCR Master Mix.

Table 3. Reaction setup

Component	Volume/reaction	Final concentration
2x Rotor-Gene Probe RT-PCR Master Mix	12.5 µl	1x
Primer A	Variable	0.8 µM
Primer B	Variable	0.8 µM
Probe	Variable	0.2 µM
Rotor-Gene RT Mix	0.25 µl	
Template RNA (added at step 4)	Variable	≤100 ng/ reaction
RNase-free water	Variable	
Total reaction volume	**25 µl**	

3. **Mix the reaction mix thoroughly, and dispense appropriate volumes into PCR tubes.**

   Keep the PCR tubes on ice.

4. **Add template RNA (≤100 ng/reaction) to the individual PCR tubes containing the reaction mix.**

5. **Program the Rotor-Gene cycler according to the program outlined in Table 4 (page 18) and Figures 3–5 (pages 19–21).**

   Data acquisition should be performed during the combined annealing/ extension step.

Table 4. Cycling conditions

Step	Time	Temperature	Additional comments
Reverse transcription	10 min	50°C	
PCR initial activation step	5 min	95°C	HotStarTaq *Plus* DNA Polymerase is activated by this heating step
Two-step cycling			
Denaturation	5 s	95°C	
Combined annealing/ extension	10 s	60°C	Perform fluorescence data collection
Number of cycles	35–40		The number of cycles depends on the amount of template RNA

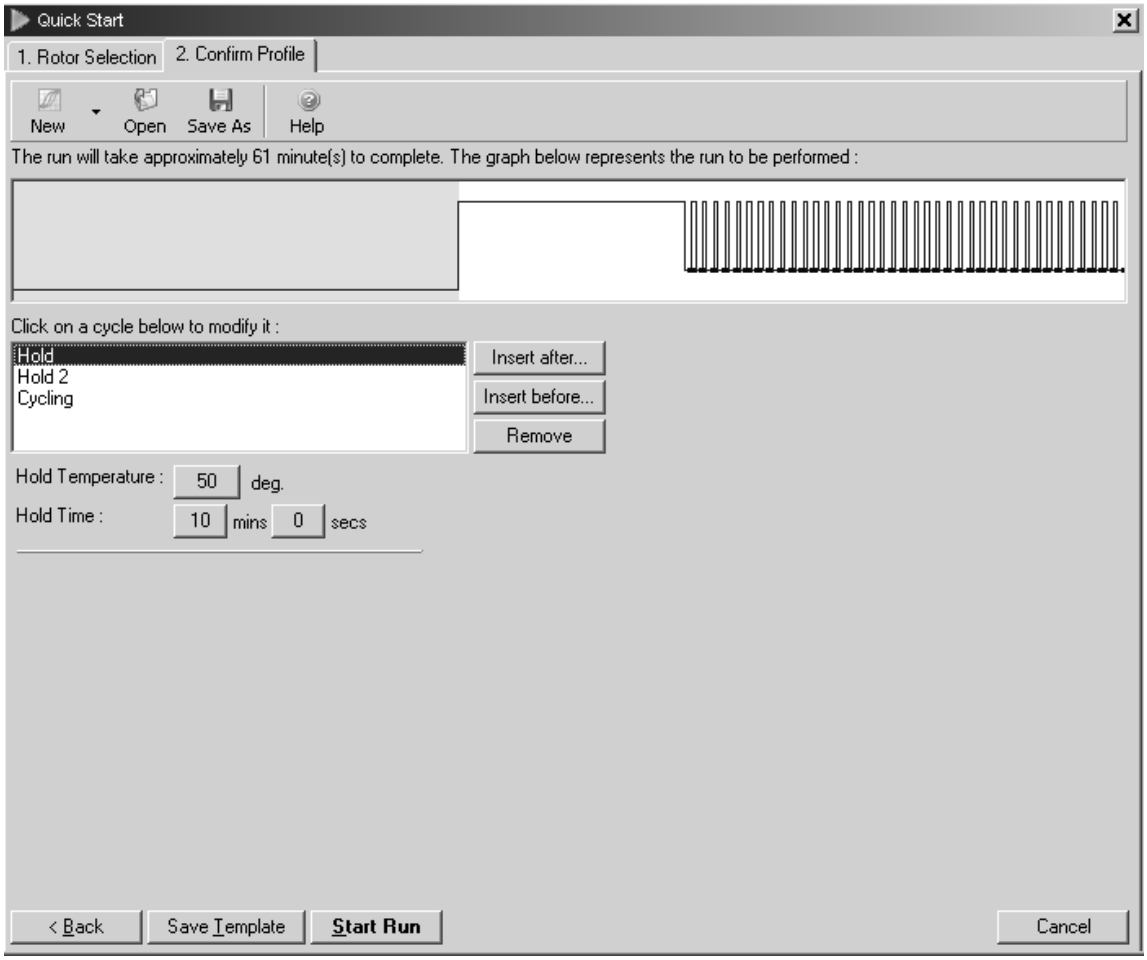

Figure 3. Reverse transcription. Before starting PCR, reverse transcription must be carried out. Reactions are incubated at 50°C for 10 min.

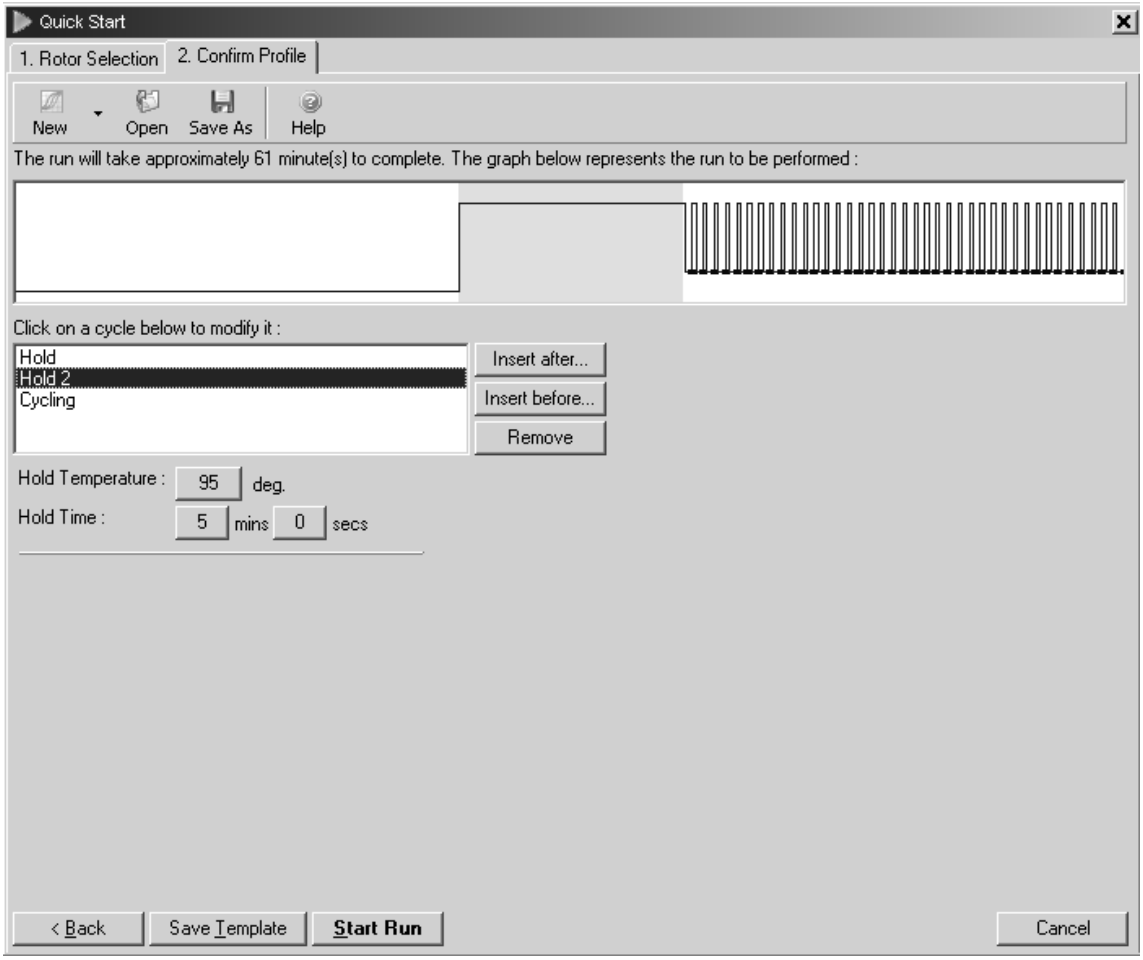

Figure 4. PCR initial activation step. After reverse transcription is completed, PCR can be carried out. PCR requires an initial incubation at 95°C for 5 min.

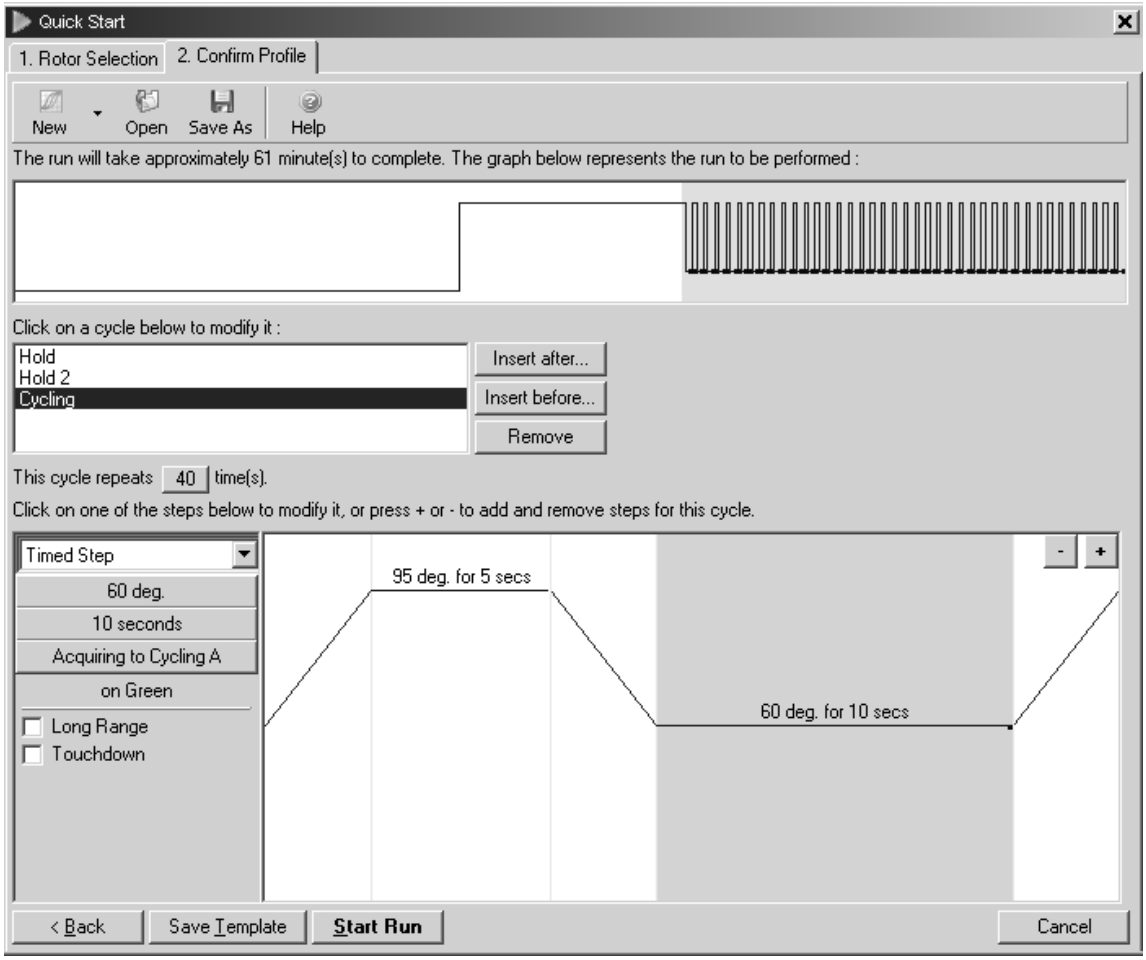

Figure 5. Two-step cycling. PCR requires 35–40 cycles. Each cycle is comprised of 2 steps: 95°C for 5 s (denaturation step) and 60°C for 10 s (annealing/extension step).

6. Place the PCR tubes in the Rotor-Gene cycler, and start the cycling program.

Troubleshooting Guide

This troubleshooting guide may be helpful in solving any problems that may arise. For more information, see also the Frequently Asked Questions page at our Technical Support Center: www.qiagen.com/FAQ/FAQList.aspx. The scientists in QIAGEN Technical Services are always happy to answer any questions you may have about either the information and protocols in this handbook or sample and assay technologies (for contact information, see back cover or visit www.qiagen.com).

	Comments and suggestions
No signal, or one or more signals detected late in PCR	
a) Wrong cycling conditions	Always start with the optimized cycling conditions specified in the protocols. Be sure that the cycling conditions include the initial step for activation of HotStarTaq *Plus* DNA Polymerase (95°C for 3 or 5 min), and the specified times for denaturation and annealing/extension. If performing one-step RT-PCR, be sure that the cycling conditions include the RT step (50°C for 10 min) prior to the HotStarTaq *Plus* DNA Polymerase activation step.
b) HotStarTaq *Plus* DNA Polymerase not activated	Ensure that the cycling program includes the HotStarTaq *Plus* DNA Polymerase activation step (3 or 5 min at 95°C) as described in the protocols.
c) Pipetting error or missing reagent	Check the concentrations and storage conditions of the reagents, including primers, probes, and template nucleic acid.* Repeat the PCR.
d) Wrong or no detection step	Ensure that fluorescence detection takes place during the combined annealing/extension step.

* For details, refer to "Guidelines for real-time PCR and RT-PCR" at www.qiagen.com/resources/info.

	Comments and suggestions
e) Primer or probe concentration not optimal	Use optimal primer concentrations. For TaqMan probes, use each primer at 0.4 µM (for PCR and two-step RT-PCR) or 0.8 µM (for one-step RT-PCR). In most cases, a probe concentration of 0.2 µM gives satisfactory results. Check the concentrations of primers and probes by spectrophotometry.* If using a commercial probe-based assay (e.g., TaqMan Gene Expression Assays), the final concentration in reactions should be 1x, as recommended by the supplier.
f) Problems with starting template	Check the concentration, storage conditions, and quality of the starting template.* If necessary, make new serial dilutions of template nucleic acid from the stock solutions. Repeat the PCR using the new dilutions.
g) Insufficient amount of starting template	Increase the amount of template, if possible. Ensure that sufficient copies of the target nucleic acids are present in your sample.
h) Insufficient number of cycles	Increase the number of cycles.
i) Reaction volume too high	We recommend a final reaction volume of 25 µl.
j) PCR product too long	For optimal results, PCR products should be between 100 and 150 bp. PCR products should not be outside the range of 70–300 bp.
k) Primer design not optimal	Check for PCR products by gel electrophoresis. If no specific PCR products are detected, review the primer design guidelines.*

* For details, refer to "Guidelines for real-time PCR and RT-PCR" at www.qiagen.com/resources/info.

	Comments and suggestions
l) Probe design not optimal	If the amplification reaction itself was successful (this can be checked by gel electrophoresis analysis of the PCR products), there may be a problem with the probe. Review the probe design guidelines.*
m) Wrong dye channel chosen	Ensure that the correct dye channel is chosen for the reporter dye.
n) PCR annealing temperature too high	Decrease annealing temperature in steps of 2°C.
o) PCR annealing temperature too low	Increase annealing temperature in steps of 2°C.
p) No detection activated	Check that fluorescence detection was activated in the cycling program.
q) Probe synthesis not optimal	Check the quality of TaqMan probes by incubation with DNase I. A correctly synthesized probe, containing both fluorophore and quencher, will show a significant increase in fluorescence after DNase I incubation.
r) Primers degraded	Check for possible degradation of primers on a denaturing polyacrylamide gel.
s) **Two-step RT-PCR only**: Volumes of RT reaction added were too high	High volumes of RT reaction added to the PCR may reduce amplification efficiency and the linearity of the reaction. Generally, the volume of undiluted RT reaction added should not exceed 10% of the final PCR volume.
t) **One-step RT-PCR**: RT step not performed	Ensure that the cycling program includes the RT step (10 min at 50°C) as described in the protocols.
No linearity in ratio of C_T value to log of the template amount	
a) Template amount too high	Do not exceed maximum recommended amounts of template.
b) Template amount too low	Increase template amount, if possible.

* For details, refer to "Guidelines for real-time PCR and RT-PCR" at www.qiagen.com/resources/info.

	Comments and suggestions
c) **Two-step RT-PCR only**: Volumes of RT reaction added were too high	High volumes of RT reaction added to the PCR may reduce amplification efficiency. Generally, the volume of undiluted reverse-transcription reaction added should not exceed 10% of the final PCR volume. If you need to use a large volume of reverse-transcription reaction as template, determine the maximum acceptable volume for the assay being carried out.
Increased fluorescence or C_T value for "No Template" control	
a) Contamination of reagents	Discard all the components of the assay (e.g., master mix, primers, and probes). Repeat the assay using new components.
b) Contamination during reaction setup	Take appropriate precautions during reaction setup, such as using aerosol-barrier pipet tips.
c) Minimal probe degradation, leading to sliding increase in fluorescence	Check the amplification plots, and adjust the threshold settings.
High fluorescence in "No Reverse Transcription" control	
Contamination of RNA sample with genomic DNA	Design primers and/or probes that span exon-exon boundaries, so that only cDNA targets can be amplified and detected. Alternatively, treat the RNA sample with DNase to digest the contaminating genomic DNA. If carrying out real-time two-step RT-PCR, perform reverse transcription with the QuantiTect Reverse Transcription Kit, which provides cDNA synthesis with integrated genomic DNA removal.

Ordering Information

Product	Contents	Cat. no.
Rotor-Gene Probe PCR Kit (400)	For 400 x 25 µl reactions: 3 x 1.7 ml 2x Master Mix, 2 x 2 ml RNase-Free Water	204374
Rotor-Gene Probe RT-PCR Kit (400)	For 400 x 25 µl reactions: 3 x 1.7 ml 2x Master Mix, 100 µl RT Mix, 2 x 2 ml RNase-Free Water	204574
Accessories		
QuantiTect Reverse Transcription Kit — for fast cDNA synthesis for sensitive real-time two-step RT-PCR		
QuantiTect Reverse Transcription Kit (50)*	For 50 x 20 µl reactions: gDNA Wipeout Buffer, Quantiscript® Reverse Transcriptase, Quantiscript RT Buffer, RT Primer Mix, and RNase-Free Water	205311
FastLane Cell cDNA Kit — for high-speed preparation of cDNA without RNA purification for use in real-time RT-PCR		
FastLane Cell cDNA Kit (50)	Buffer FCW, Buffer FCP, and components for 50 x 20 µl reverse-transcription reactions (gDNA Wipeout Buffer, Quantiscript Reverse Transcriptase, Quantiscript RT Buffer, RT Primer Mix, and RNase-Free Water)	215011
Related products		
Rotor-Gene SYBR Green PCR Kit — for fast real-time PCR and two-step RT-PCR using SYBR Green I on Rotor-Gene cyclers		
Rotor-Gene SYBR Green PCR Kit (400)	For 400 x 25 µl reactions: 3 x 1.7 ml 2x Master Mix, 2 x 2 ml RNase-Free Water	204074
Rotor-Gene SYBR Green RT-PCR Kit — for fast real-time one-step RT-PCR using SYBR Green I on Rotor-Gene cyclers		
Rotor-Gene SYBR Green RT-PCR Kit (400)	For 400 x 25 µl reactions: 3 x 1.7 ml 2x Master Mix, 100 µl RT Mix, 2 x 2 ml RNase-Free Water	204174

* Trial-size kit and larger kit available; please inquire.

Product	Contents	Cat. no.
Rotor-Gene Multiplex PCR Kit — for fast multiplex real-time PCR and two-step RT-PCR on Rotor-Gene cyclers		
Rotor-Gene Multiplex PCR Kit (80)	For 80 x 25 µl reactions: 1 ml 2x Master Mix, 2 ml RNase-Free Water	204772
Rotor-Gene Multiplex PCR Kit (400)	For 400 x 25 µl reactions: 3 x 1.7 ml 2x Master Mix, 2 x 2 ml RNase-Free Water	204774

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at www.qiagen.com or can be requested from QIAGEN Technical Services or your local distributor.

Visit www.qiagen.com/geneXpression to find out more about standardized solutions for gene expression analysis — from RNA preparation to real-time RT-PCR

Notes

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