November 2010

HiPerFect® HTS Reagent Handbook

For high-throughput transfection of eukaryotic cells with siRNA

Sample & Assay Technologies

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Contents

Kit Contents

HiPerFect HTS Reagent	0.1 ml	2 x 1 ml	6 x 1 ml
Catalog no.	**301802**	**301806**	**301807**
Transfections per kit (number of 96-well plates)	**4–10 plates**	**80–200 plates**	**240–600 plates**
HiPerFect HTS Reagent	0.1 ml	2 x 1 ml	6 x 1 ml
Protocol Card	2*	2*	2*

* Two Protocol Cards are provided with the kit detailing transfection protocols for 96-well plates or 384-well plates. This handbook provides more detailed information. To download QIAGEN handbooks, visit www.qiagen.com/handbooks.

Storage

HiPerFect HTS Transfection Reagent is supplied as a ready-to-use solution and is shipped at ambient temperature without loss of stability. However, it should be stored at 2–8°C upon arrival. HiPerFect HTS Transfection Reagent does not need to be stored on ice during the transfection procedure.

Product Use Limitations

HiPerFect HTS Transfection Reagent is intended for molecular biology applications. This product is 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).

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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 material safety data sheets (MSDSs). These are available online in convenient and compact PDF format at www.qiagen.com/Support/MSDS.aspx where you can find, view, and print the MSDS for each QIAGEN kit and kit component.

24-hour emergency information

Emergency medical information in English, French, and German can be obtained 24 hours a day from:

Poison Information Center Mainz, Germany

Tel: +49-6131-19240

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of HiPerFect HTS Reagent is tested against predetermined specifications to ensure consistent product quality.

Introduction

The technique of RNAi has revolutionized research in the fields of functional genomics and drug discovery. Increasingly, researchers are using high-throughput techniques for RNAi analysis to identify genes whose aberrant regulation affects a biological process involved in disease, such as cell proliferation, tumor suppression, or phosphorylation cascades. Alternatively, research in pathway analysis could focus on the identification of genes involved in the regulation of a specific protein by, for example, observing protein translocation.

Efficient transfection of siRNA is critical for effective gene silencing. High-throughput transfection conditions must be highly optimized and standardized to minimize artifacts and maximize reproducibility. Transfections should be carried out using low siRNA concentrations and a reagent that ensures low cytotoxicity. HiPerFect HTS Reagent has been developed for efficient transfection of cells even when low siRNA concentrations are used. In addition, cells transfected using HiPerFect HTS Reagent show high cell viability and low cytotoxicity providing reliable and accurate results.

Principle and procedure

HiPerFect HTS Reagent has been developed especially for use in high-throughput RNAi screening experiments. HiPerFect HTS Reagent provides robust and reliable transfection ensuring consistency and reproducibility in experiments involving large numbers of transfections. The protocol is designed to minimize consumable use and pipetting steps, saving resources and time as well as facilitating standardization. The straightforward reverse-transfection protocol is a rapid, convenient method for high-throughput experiments.

Description of protocols

This handbook contains 2 protocols for transfection using HiPerFect HTS Reagent. The protocols describe reverse transfection of adherent cells in 96-well plates (page 13) or 384-well plates (page 15). In these protocols, cells are seeded and transfected on the same day. siRNA is spotted into wells followed by the addition of HiPerFect HTS Reagent. After complex formation, cells are added to the wells.

Reagents to Be Supplied by User

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, consult the appropriate material safety data sheets (MSDSs), available from the product supplier.

- Culture medium
- siRNA of interest. We recommend FlexiPlate siRNA, which provides maximum flexibility to select siRNAs targeting human and mouse genes, controls, scales, and plate layout. The GeneGlobe® Web portal allows convenient ordering (www.qiagen.com/GeneGlobe).

Important Notes

Calculating concentrations of siRNA

Approximate values for a double-stranded, 21 nt siRNA molecule:

- 20 µM siRNA is equivalent to approximately 0.25 µg/µl
- The molecular weight of a 21 nt siRNA is approximately 13–15 µg/nmol

Optimizing siRNA transfection

To achieve the best results in siRNA transfection of adherent cells, we recommend optimizing the following parameters.

Amount of siRNA

The amount of siRNA used is critical for efficient transfection and gene silencing. The recommended starting concentration for transfection of siRNA in 96-well and 384-well plates is 5 nM. However, the amount of siRNA required for optimal performance may vary, depending on the cell line and gene target. Suggested starting concentrations for a range of cell lines are shown in Tables 1 and 2. An example pipetting scheme for optimizing siRNA transfection of adherent cells in 96-well plates is shown in Table 3 (page 10).

Ratio of HiPerFect HTS Reagent to siRNA

The ratio of HiPerFect HTS Reagent to siRNA should be optimized for every new cell type and siRNA combination used. As a starting point for optimization, we recommend 5 nM siRNA and 0.25 µl HiPerFect HTS Reagent (96-well plates) or 5 nM siRNA and 0.1 µl HiPerFect HTS Reagent (384-well plates). However, the amount of HiPerFect HTS Transfection Reagent and siRNA required for optimal performance may vary, depending on the cell line and gene target. Suggested starting amounts for a range of cell lines are shown in Tables 1 and 2.

To optimize siRNA transfection in 96-well plates, prepare separate transfection mixtures as shown in the example in Table 3. Please note that Table 3 is intended only as a guideline for starting amounts of siRNA and reagent. These amounts worked well as a starting point for transfection optimization in cell lines tested using HiPerFect HTS Reagent.

Table 1. Starting points for optimizing transfection of adherent cell lines in 96-well plates

Cell line	siRNA concentration (nM/well)	HiPerFect HTS Reagent (µl/well)
A549	10	0.5
AGS	25	0.25
HeLa	2.5	0.1
HepG2	5	0.25
MCF-7	25	0.25
NIH-3T3	25	0.3
293	10	0.25

Table 2. Starting points for optimizing transfection of adherent cell lines in 384-well plates

Cell line	siRNA concentration (nM/well)	HiPerFect HTS Reagent (µl/well)
A549	5	0.25
AGS	10	0.25
HeLa	25	0.05
HepG2	5	0.1
Huh-7	25	0.25
MCF-7	5	0.1
NIH-3T3	10	0.1
293	10	0.25

Table 3. Pipetting scheme for optimizing transfection of adherent cells in 96-well plates*

Amount (conc.) of siRNA	25 ng (10 nM)	25 ng (10 nM)	25 ng (10 nM)
Volume of HiPerFect HTS Reagent	0.1 µl	0.25 µl	0.5 µl
Amount (conc.) of siRNA	12.5 ng (5 nM)	**12.5 ng (5 nM)**	12.5 ng (5 nM)
Volume of HiPerFect HTS Reagent	0.1 µl	**0.25 µl**	0.5 µl
Amount (conc.) of siRNA	2.5 ng (1 nM)	2.5 ng (1 nM)	2.5 ng (1 nM)
Volume of HiPerFect HTS Reagent	0.1 µl	0.25 µl	0.5 µl

* Amounts given are per well of a 96-well plate. siRNA concentrations shown in brackets refer to the final siRNA concentrations in each well.

Cell density at transfection

The optimal cell confluency for transfection should be determined for every new cell type to be transfected and kept constant in future experiments. This is achieved by counting cells before seeding. This ensures that the cell density is not too high and that the cells are in optimal physiological condition at transfection. The recommended number of cells to seed for each format is provided in the protocol.

Transfection in multiwell plates — preparing a master mix

When performing transfection in multiwell plates, prepare a master mix of transfection reagent and culture medium for distribution into plate wells.

- Calculate the required volumes of each component and the total volume before you prepare the master mix.
- Prepare 10% more master mix than is required to allow for pipetting errors (i.e., for a 96-well plate, prepare enough master mix for 106 wells).
- Add and mix the components of the master mix according to the instructions in the protocol.
- Use a repeat pipet to distribute the master mix.

Performing appropriate RNAi control experiments

It is important to perform suitable control experiments so that results can be correctly interpreted. A full range of control siRNAs is available at www.qiagen.com/AllStars. We recommend the following control experiments.

Positive control siRNA

Positive control siRNA is known to provide high knockdown of its target gene. For example, AllStars Cell Death Control siRNAs (cat. no. 1027298 for human; cat. no. SI04939025 for mouse and rat) provides high knockdown of ubiquitous human cell survival genes which results in a high degree of cell death which is visible by light microscopy. These siRNAs can be used as positive controls. They can also be used for transfection optimization, for example, when establishing RNAi in a new cell line.

A positive control is used to establish that the experimental set up for transfection and knockdown analysis is working optimally. An siRNA that knocks down a gene resulting in the phenotypic effect under study may also be used as a positive control to ensure that the phenotypic assay is working optimally. A positive control siRNA should be transfected in every RNAi experiment.

Negative control siRNA

A negative control siRNA should be a nonsilencing siRNA with no homology to any known mammalian gene, such as AllStars Negative Control siRNA (5 nmol) from QIAGEN (cat. no. 1027280). AllStars Negative Control siRNA is the most thoroughly tested and validated negative control available. It has been shown to provide minimal nonspecific effects on gene expression and phenotype and is incorporated into RISC. For more details and to view data, visit www.qiagen.com/AllStars. Transfection of negative control siRNA is used to determine whether changes in phenotype or gene expression are nonspecific. A negative control siRNA should be transfected in every RNAi experiment.

Mock transfection control

Mock-transfected cells go through the transfection process without addition of siRNA (i.e., cells are treated with transfection reagent only). This control is used to determine any nonspecific effects that may be caused by the transfection reagent or process.

Untransfected cells control

Gene expression analysis should be carried out on cells that have not been treated to allow measurement of the normal, basal level of gene expression. Results from untreated cells can be used for comparison with results from all other samples. Untreated cells should be analyzed in every RNAi experiment.

Additional siRNAs for phenotype confirmation

A phenotypic effect caused by knockdown of a gene must be confirmed using at least one additional siRNA targeted against a different area of the mRNA.

Monitoring gene silencing at the mRNA or protein level

Gene silencing can be monitored at either the mRNA or the protein level. Protein analysis can be performed using western blotting, immunofluorescence, or FACS® analysis. More information about protein analysis and a protocol for western blotting can be found at www.qiagen.com/literature/BenchGuide. The AllPrep RNA/Protein Kit (cat. no. 80404) allows simultaneous purification of RNA and protein from the same sample for streamlined downstream analysis after knockdown. Protein analysis is the most comprehensive way of showing that a gene has been downregulated. However, it may not always be possible to perform protein analysis (e.g., if antibodies for the protein of interest are not available for western blotting). Silencing is usually monitored at the mRNA level by real-time RT-PCR, microarray analysis, or northern blotting. Information about working with RNA and a northern blotting protocol are available at www.qiagen.com/literature/BenchGuide. Quantitative, real-time RT-PCR is an easy and routinely used method to monitor gene silencing at the mRNA level. QuantiTect® Primer Assays are bioinformatically validated primer sets that are available for every human, mouse, rat, dog, drosophila, chicken, and Arabidopsis gene. QuantiTect Primer Assays are used in combination with QuantiTect or QuantiFast® SYBR® Green Kits for sensitive and specific one-step or two-step real-time RT-PCR using SYBR Green detection. Expression data should be compared with levels of a housekeeping gene, such as GAPDH, to normalize for variable amounts of RNA in different samples.

Protocol: Reverse Transfection of Adherent Cells with siRNA in 96-Well Plates

This protocol is provided as a starting point for optimization of siRNA transfection of adherent cells in a single well of a 96-well plate using HiPerFect HTS Reagent. Starting points for optimizing transfection in individual cell lines are listed in Table 1 (page 9). In this protocol, cell plating and transfection are performed on the same day.

Note: Transfection in 96-well plates can also be performed using a Fast-Forward Protocol, if desired. In this Reverse-Transfection Protocol, complex formation takes place in the plate wells first and then cells are added on top of the complexes. In the Fast-Forward Protocol, cells are added to plate wells followed by complexes. To perform a Fast-Forward Protocol, simply change the order in which cells and complexes are added to the plate (steps 1–4). However, we recommend using this Reverse-Transfection Protocol when working with 96-well plates because it is quicker, has fewer pipetting steps, and uses fewer materials (the Fast-Forward Protocol requires two plates, one for complex formation and one for adding complexes on top of cells; the Reverse-Transfection Protocol requires only one plate).

Important points before starting

- Cells should be in optimal physiological condition at the time of transfection. The optimal amount of cells seeded depends on the cell type and time of analysis.
- If siRNA has been ordered from QIAGEN, it is delivered lyophilized and must be resuspended prior to transfection. To resuspend, follow the instructions provided with the siRNA.

Procedure

1. **Spot 12.5 ng siRNA in 1–3 µl siRNA Suspension Buffer/RNase-free water into a single well of a 96-well plate (this will give a final siRNA concentration of 5 nM after addition of cells to complexes in step 4).**

   Note: After this step, siRNA may be stored at –20°C for long time periods. siRNA may be stored in solution or may be dried in the plate at room temperature (15–25°C) before storage.

   Note: If preferred, siRNA can be spotted in 25 µl of siRNA Suspension Buffer/RNase-free water into each well. In this case, 150 µl culture medium (containing 1–5 x 10^4 cells) should be added in step 4.

2. **Add 0.25 µl HiPerFect HTS Reagent to 24.75 µl culture medium without serum. Add the diluted HiPerFect HTS Reagent to the prespotted siRNA.**

Note: To ensure accurate pipetting, diluted HiPerFect HTS Reagent should be prepared in a larger volume for use in multiple wells. Then add 25 µl of the dilution to a single well.

IMPORTANT: The amount of HiPerFect HTS Transfection Reagent and siRNA required for optimal performance may vary, depending on the cell line and gene target. For suggested starting points for optimization of siRNA to HiPerFect HTS Reagent ratio, see Table 1, page 9.

3. **Incubate for 5–10 min at room temperature (15–25°C) to allow formation of transfection complexes.**
4. **Seed 0.25–1 x 10^4 cells in 175 µl of an appropriate culture medium (containing serum and antibiotics) into the well, on top of the siRNA–HiPerFect HTS Reagent transfection complexes.**
5. **Incubate the cells with the transfection complexes under their normal growth conditions and monitor gene silencing after an appropriate time (e.g., 6–72 h after transfection, depending on experimental setup). Change the medium as required.**

   Note: The optimal incubation time for gene silencing analysis depends on cell type, the gene targeted, and the method of analysis. This can be determined by performing a time-course experiment.

Protocol: Reverse Transfection of Adherent Cells with siRNA in 384-Well Plates

This protocol is provided as a starting point for optimization of siRNA transfection of adherent cells in a single well of a 384-well plate using HiPerFect HTS Reagent. Starting points for optimizing transfection in individual cell lines are listed in Table 2 (page 9). In this protocol, cell plating and transfection are performed on the same day.

Important points before starting

- Cells should be in optimal physiological condition at the time of transfection. The optimal amount of cells seeded depends on the cell type and time of analysis.
- If siRNA has been ordered from QIAGEN, it is delivered lyophilized and must be resuspended prior to transfection. To resuspend, follow the instructions provided with the siRNA.

Procedure

1. **Spot 3.125 ng siRNA in 1–3 µl siRNA Suspension Buffer/RNase-free water into a single well of a 384-well plate (this will give a final siRNA concentration of 5 n*M* after addition of cells to complexes in step 4).**

   Note: After this step, siRNA may be stored at –20°C for long time periods. siRNA may be stored in solution or may be dried in the plate at room temperature (15–25°C) before storage.

2. **Add 0.1 µl HiPerFect HTS Reagent to 9.9 µl culture medium without serum. Add the diluted HiPerFect HTS Reagent to the prespotted siRNA.**

   Note: To ensure accurate pipetting, diluted HiPerFect HTS Reagent should be prepared in a larger volume for use in multiple wells. Then add 10 µl of the dilution to a single well.

   IMPORTANT: The amount of HiPerFect HTS Transfection Reagent and siRNA required for optimal performance may vary, depending on the cell line and gene target. For suggested starting points for optimization of siRNA to HiPerFect HTS Reagent ratio, see Table 2, page 9.

3. **Incubate for 5–10 min at room temperature (15–25°C) to allow formation of transfection complexes.**
4. **Seed 1000–3000 cells in 40 µl of an appropriate culture medium (containing serum and antibiotics) into the well, on top of the siRNA–HiPerFect HTS Reagent transfection complexes.**

5. **Incubate the cells with the transfection complexes under their normal growth conditions and monitor gene silencing after an appropriate time (e.g., 6–72 h after transfection, depending on experimental setup). Change the medium as required.**

   Note: The optimal incubation time for gene silencing analysis depends on cell type, the gene targeted, and the method of analysis. This can be determined by performing a time-course experiment.

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
Low transfection efficiency	
a) Suboptimal HiPerFect HTS Reagent:siRNA ratio	Although fixed volumes of HiPerFect HTS Reagent usually work very well with a range of siRNA concentrations, it could occur that the overall charge of the complexes is negative, neutral, or strongly positive, which can lead to inefficient adsorption to the cell surface. For optimal adsorption, complexes should be weakly positive. Optimize the HiPerFect HTS Reagent to siRNA ratio or perform systematic titrations of HiPerFect HTS Reagent.
b) Suboptimal cell density	If cell density at the time of adding HiPerFect HTS Reagent–siRNA complexes is not at an optimal level, cells may not be in the optimal growth phase for transfection. This can lead to insufficient uptake of complexes into the cells or inefficient processing of the siRNA. For adherent cells, the optimal confluency for transfection of siRNA is 50–80% on the day of transfection.
c) Poor siRNA quality	siRNA should be of high quality, as impurities can reduce transfection efficiency. We recommend FlexiPlate siRNA for efficient gene silencing (www.qiagen.com/GeneGlobe).
Excessive cell death	
a) Concentration of HiPerFect HTS Reagent–siRNA complexes is too high	Decrease the amount of HiPerFect HTS Reagent–siRNA complexes added to the cells.

	Comments and suggestions
b) Cells are stressed	Avoid stressing cells with temperature shifts and long periods without medium during washing steps. It is particularly important for transfection of siRNA that the cells are in good condition. Therefore, ensure that cell density is not too low at transfection. For adherent cells, the optimal confluency for transfection is 50–80% on the day of transfection.
c) Poor siRNA quality	siRNA should be of high quality, as impurities can reduce transfection efficiency. We recommend FlexiPlate siRNA for efficient gene silencing (www.qiagen.com/GeneGlobe).
d) Key gene is silenced	If the gene targeted is important for the survival of the cell, silencing this gene may lead to cell death.

Variable transfection efficiencies in replicate experiments

a) Inconsistent cell confluencies in replicate experiments	Count cells before seeding to ensure that the same number of cells is seeded for each experiment.
b) Possible mycoplasma contamination	Mycoplasma contamination influences transfection efficiency. Variations in the growth behavior of mycoplasma-infected cells will lead to different transfection efficiencies between replicate experiments.
c) Cells have been passaged too many times	Cells that have been passaged a large number of times tend to change their growth behavior and morphology, and are less susceptible to transfection. When cells with high passage numbers are used for replicate experiments, decreased transfection efficiencies may be observed in later experiments. We recommend using cells with a low passage number (<50 splitting cycles).
d) Concentration of siRNA is too low	Increase siRNA concentration used in transfection.

	Comments and suggestions
No or very small gene silencing effect	
a) Design of siRNA suboptimal	The design of an siRNA can have a large effect on its gene silencing efficiency. We recommend FlexiPlate siRNA for efficient gene silencing (www.qiagen.com/GeneGlobe).
b) Incubation time after transfection too short	The gene silencing effect observed at the protein level is dependent on the expression level of the protein and its rate of turnover within the cell. Perform a time-course experiment to determine the optimal time point for analysis.
c) Problems with experimental design	RNAi effects may not be seen for some genes targeted with certain siRNAs in some cell types. If possible, repeat experiments using a different cell type and/or siRNA. Where possible, include both positive and negative controls in your experiments. QIAGEN offers a range of control siRNAs at www.qiagen.com/AllStars.
d) Concentration of siRNA is too low	Increase siRNA concentration used in transfection.

References

QIAGEN maintains a large, up-to-date online database of scientific publications utilizing QIAGEN products. Comprehensive search options allow you to find the articles you need, either by a simple keyword search or by specifying the application, research area, title, etc.

For a complete list of references, visit the QIAGEN Reference Database online at www.qiagen.com/RefDB/search.asp or contact QIAGEN Technical Services or your local distributor.

Ordering Information

Product	Contents	Cat. no.
HiPerFect HTS Reagent (0.1 ml)	Reagent for transfections in 4–10 ninety-six–well plates	301802
HiPerFect HTS Reagent (2 x 1 ml)	Reagent for transfections in 80–200 ninety-six–well plates	301806
HiPerFect HTS Reagent (6 x 1 ml)	Reagent for transfections in 240–600 ninety-six–well plates	301807
Related products		
FlexiPlate siRNA	Custom siRNA set for customer-specified genes and siRNA controls; minimum order 36 siRNAs; 0.1 nmol, 0.25 nmol, or 1 nmol scale; plate layout chosen by the customer at GeneGlobe	Varies*
FlexiTube siRNA	Predesigned siRNAs for human, mouse, or rat genes ; delivered in tubes	Varies*
FlexiTube GeneSolution	4 siRNAs recommended for your human or mouse gene at GeneGlobe; siRNAs delivered in tubes in 1 nmol amounts	Varies*
FlexiTube siRNA Premix	Optimized siRNA–transfection reagent mix; 0.75 nmol siRNA; provided in tubes	Varies*
AllStars Negative Control siRNA (5 nmol)	Validated siRNA with no homology to any known mammalian gene, for use as a nonsilencing control	1027280
AllStars Hs Cell Death Control siRNA (5 nmol)	Positive cell death phenotype control for human cells	1027298
AllStars Mm/Rn Cell Death Control siRNA (1 nmol)	Positive cell death phenotype control for mouse or rat cells	SI04939025

* Visit www.qiagen.com/GeneGlobe to search for and order these products.

Product	Contents	Cat. no.
RNeasy Mini Kit (50)*	50 RNeasy Mini Spin Columns, Collection Tubes (1.5 ml and 2 ml), RNase-Free Reagents and Buffers	74104
AllPrep RNA/Protein Kit (50)	50 AllPrep Mini Spin Columns, 50 RNeasy Mini Spin Columns, 50 Protein Cleanup Mini Spin Columns, Collection Tubes, RNase-Free Reagents and Buffers	80404
QuantiTect Primer Assay (200)	For 200 x 50 µl real-time PCRs: lyophilized mix of forward and reverse primers	Varies†
QuantiTect Reverse Transcription Kit (50)†	For 50 x 20 µl reverse-transcription reactions: gDNA Wipeout Buffer, Quantiscript® Reverse Transcriptase, Quantiscript RT Buffer, RT Primer Mix, RNase-Free Water	205311
QuantiFast 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	204154
QuantiFast 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	204054

* Also available in a larger size and in micro, midi, maxi, and 96-well formats; please inquire.

† Visit www.qiagen.com/GeneGlobe to search for and order these products.

‡ Also available in larger kit sizes; please inquire.

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Canada ■ Orders 800-572-9613 ■ Fax 800-713-5951 ■ Technical 800-DNA-PREP (800-362-7737)

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France ■ Orders 01-60-920-926 ■ Fax 01-60-920-925 ■ Technical 01-60-920-930 ■ Offers 01-60-920-928

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