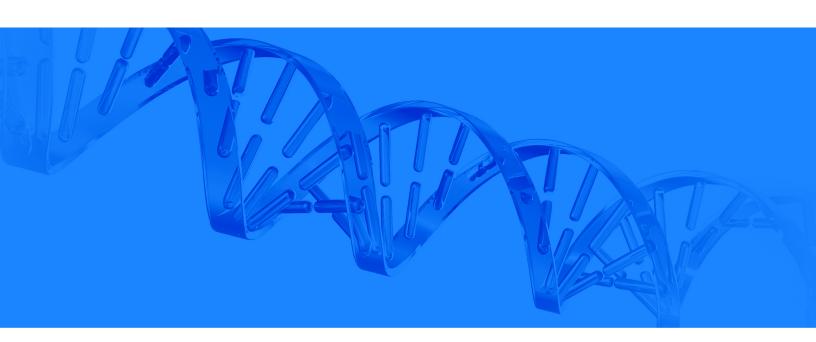


Protein Expression Sample Preparation



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Safety information

Note: For general safety information, see this Preface and Appendix C, "Safety" on page 15. When a hazard symbol and hazard type appear by a chemical name or instrument hazard, see the "Safety" Appendix for the complete alert on the chemical or instrument.

Safety alert words

Four safety alert words appear in Applied Biosystems user documentation at points in the document where you need to be aware of relevant hazards. Each alert word—**IMPORTANT**, **CAUTION**, **WARNING**, **DANGER**—implies a particular level of observation or action, as defined below:

IMPORTANT! – Indicates information that is necessary for proper instrument operation, accurate chemistry kit use, or safe use of a chemical.



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.



DANGER! – Indicates an imminently hazardous situation that, if not avoided, will result in death or serious injury. This signal word is to be limited to the most extreme situations.

MSDSs

The MSDSs for any chemicals supplied by Applied Biosystems or Ambion are available to you free 24 hours a day. For instructions on obtaining MSDSs, see "MSDSs" on page 17.

IMPORTANT! For the MSDSs of chemicals not distributed by Applied Biosystems or Ambion contact the chemical manufacturer.

How to use this guide

Text conventions

This guide uses the following conventions:

- **Bold** text indicates user action. For example:
 - Type 0, then press **Enter** for each of the remaining fields.
- *Italic* text indicates new or important words and is also used for emphasis. For example:
 - Before analyzing, *always* prepare fresh matrix.
- A right arrow symbol (▶) separates successive commands you select from a drop-down or shortcut menu. For example:
 - Select File ▶ Open ▶ Spot Set.

Right-click the sample row, then select View Filter > View All Runs.

User attention words

Two user attention words appear in Applied Biosystems user documentation. 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, accurate chemistry kit use, or safe use of a chemical.

How to obtain support

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

www.appliedbiosystems.com

At the Applied Biosystems web site, you can:

- Access worldwide telephone and fax numbers to contact Applied Biosystems Technical Support and Sales facilities.
- Search through frequently asked questions (FAQs).
- Submit a question directly to Technical Support.
- Order Applied Biosystems user documents, MSDSs, certificates of analysis, and other related documents.
- · Download PDF documents.
- · Obtain information about customer training.
- Download software updates and patches.

Protein Expression Sample Preparation Kit

Product overview

Protein Expression Sample Preparation Kit The Protein Expression Sample Preparation Kit supplies reagents used to prepare mammalian cell lysates for use with TaqMan[®] Protein Expression Assay Kits. The assays allow for the detection and relative quantitation of protein targets in a variety of cell culture samples by a real-time PCR instrument.

About this protocol

The Protein Expression Sample Preparation Protocol describes the:

- Protein Expression Sample Preparation Kit
- Materials that are required for sample preparation
- Procedures and guidelines for preparing samples for relative quantitation using the TaqMan® Protein Expression Assay Kits

Audience

This protocol is intended for novice and experienced laboratory personnel who perform experiments using the Protein Expression Assay Sample Preparation Kit.

Assumptions

This protocol assumes that you have a working knowledge of general techniques for handling cell cultures and harvesting cells for lysis.

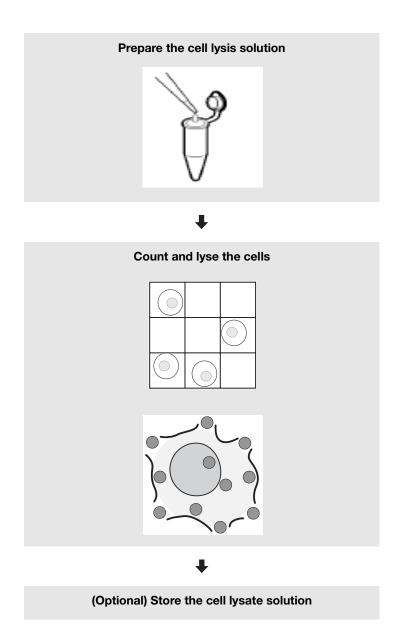
How to order materials

For information on how to order the Protein Expression Sample Preparation Kit and related materials, see Appendix A, "Ordering Information" on page 7.

For more information

For information about the TaqMan Protein Expression Assay Kits, see "Documentation and Support" on page 25.

Workflow



Prepare the cell lysis solution

In this step, you prepare the solution that you use to lyse the cells. The cell lysis solution is a mixture of protease and phosphatase inhibitor cocktails and the Cell Lysis Reagent, a buffered detergent solution.

For the following hazard, see the complete safety alert descriptions in Appendix C on page 15.



WARNING! CHEMICAL HAZARDS. Cell Lysis Reagent (2×), Protease Inhibitor Cocktail Set I (100×), Phosphatase Inhibitor Cocktail Set II (100×).

IMPORTANT! Prepare the cell lysis solution immediately before you lyse the cells. Discard any unused solution after 2 hours.

- **1.** Put the following reagents on ice:
 - Cell Lysis Reagent, 1 mL tube
 - Protease inhibitor such as the Calbiochem[®] Protease Inhibitor Cocktail Set I, 1 mL tube
 - Phosphatase inhibitor such as the Calbiochem[®] Phosphatase Inhibitor Cocktail Set II, 1 mL tube

Note: Calbiochem® Protease Inhibitor Cocktail Set I and the Calbiochem® Phosphatase Inhibitor Cocktail Set II are not included in the Protein Expression Sample Preparation Kit. The Calbiochem® Phosphatase Inhibitor Cocktail Set II is supplied as a 100× solution in water.

- **2.** If necessary, reconstitute the Calbiochem Protease Inhibitor Cocktail Set I to a 100X stock solution while the other reagents thaw:
 - **a.** Add 1 mL molecular biology grade nuclease-free H₂O to the vial containing the Calbiochem[®] Protease Inhibitor Cocktail Set I (lyophilized powder).
 - **b.** Briefly vortex the Calbiochem® Protease Inhibitor Cocktail Set I.
 - c. Place the vial on ice.

Note: You can store the reconstituted Protease Inhibitor Cocktail solution at -20 °C for repeated use.

3. Combine the reagents shown in a tube of appropriate volume.

Cell lysis solution	Volume (μL) [‡]
Cell Lysis Reagent, 2X	500
Calbiochem® Phosphatase Inhibitor Cocktail Set II, 100X	5
Calbiochem® Protease Inhibitor Cocktail Set I, 100X	5
Total Volume	510

- ‡ These volumes are sufficient to prepare 1 mL of cell lysate.
- **4.** Touch vortex the tube to mix the contents, then briefly centrifuge the tube.
- **5.** Put the cell lysis solution on ice.
- **6.** Proceed immediately to "Count and lyse the cells" on page 5.

IMPORTANT! Perform cell lysis immediately after preparing the cell lysis solution. Discard any unused solution after 2 hours.

Count and lyse the cells

About cell sample concentration

TaqMan[®] Protein Expression relative quantitation experiments are highly dependent on the accurate quantitation of cells in the test samples. For the TaqMan[®] Protein Expression Assay Kits, Applied Biosystems recommends that you prepare:

- 500 cells/µL per sample
- The same number of cells for all samples

TaqMan Protein Expression Assays are optimized for mammalian cell cultures at a starting sample concentration of 500 cells/ μ L, which is further diluted during assay setup. However, the sample input ranges can vary depending on the concentration of the target protein in the cells of each test sample.

Applied Biosystems recommends that you perform a cell-count analysis of all test samples using hemocytometry.

Note: For best results, perform the isolation, counting, and lysis steps for one sample at a time, instead of performing the steps for several samples at once.

Isolate and count the cells

For the following hazard, see the complete safety alert descriptions in Appendix C on page 15.



WARNING! CHEMICAL HAZARD. Cell Resuspension Buffer, Phosphate Buffered Saline (PBS, 1×).

- 1. Collect the sample cells using your laboratory protocol.
- 2. Count the cells, using hemocytometry.
- **3.** Put the following reagents on ice:
 - Cell Resuspension Buffer
 - Cell lysis solution (See page 4.)
- **4.** Centrifuge the cells at $500 \times g$ for 4 minutes, then rinse two times with PBS (without Ca⁺⁺ or Mg⁺⁺) to remove residual media or trypsin components.
- **5.** Centrifuge the cells again, then carefully remove all of the supernatant. Put the tube on ice.

Lyse the cells

In this step, you resuspend the cells in Cell Resuspension Buffer before adding the lysis reagent.

- 1. Resuspend the cell pellet by adding sufficient ice-cold Cell Resuspension Buffer to achieve a concentration of 1000 cells/ μ L. If the total volume is > 500 μ L, then transfer 500 μ L of the resuspended cells to a fresh tube. Use the fresh tube of cells for steps 2 and 3.
- 2. Add a volume of cell lysis solution equal to the volume in the tube to achieve a concentration of 500 cells/ μ L.
 - For example, adding 100 μ L of cell lysis solution to a 100 μ L suspension of cells at 1,000 cells/ μ L would generate 200 μ L of lysate at 500 cells/ μ L.
- **3.** Mix the solution thoroughly by gently pipetting up and down several times, avoiding excess detergent foaming.

(Optional) Store the lysates

Note: Applied Biosystems recommends that you perform the TaqMan[®] Protein Expression Assay soon after you prepare the cell lysates.

If you do not proceed immediately to the TaqMan[®] Protein Expression Assay, divide the cell lysates into volumes of about 50 μ L, then store the lysates at \leq -50 °C (e.g., -70 °C).

Troubleshooting

For detailed troubleshooting information for TaqMan protein expression experiments, see the *Real-Time PCR Systems Protein Expression Assays Chemistry Guide* (PN 4405780).

Ordering Information

Required materials

Available sample preparation kits

Table 1 shows the Protein Expression Sample Preparation Kit currently available from Applied Biosystems. You can order sample preparation kits and assays from the Applied Biosystems website as described in "Ordering from the Applied Biosystems website" on page 11.

Table 1 Protein Expression Sample Preparation Kit

Kit	Contents	Samples [‡]	Storage conditions	Part number
Protein Expression Sample Preparation Kit (PN 4405443)	Cell Lysis Reagent, 2X (3 × 1 mL)	25	2 to 8 °C	4405374
	Cell Resuspension Buffer (25 mL)			4405377

[‡] These volumes of reagent are sufficient to prepare 240 µL of cell lysate per sample for 25 samples (or 6 mL of cell lysate).

Required user-supplied materials

Obtain the materials shown in Table 2 for sample preparation. Unless otherwise indicated, all materials are available from major laboratory suppliers (MLS).

Table 2 Required materials

Material	Source [‡]
Calbiochem® Protease Inhibitor Cocktail Set I (Calbiochem PN 539131)	EMD Chemicals Inc.
Calbiochem® Phosphatase Inhibitor Cocktail Set II (Calbiochem PN 524625)	
Nuclease-free water (no diethyl pyrocarbonate [DEPC])	MLS
Phosphate-buffered saline (PBS) without Ca ⁺⁺ or Mg ⁺⁺ , pH 7.4	MLS

[‡] For the MSDS of any chemical not distributed by Applied Biosystems, contact the chemical manufacturer. Before handling any chemicals, refer to the MSDS provided by the manufacturer, and observe all relevant precautions.

Related materials and equipment

Available assays

Table 3 shows the TaqMan® Protein Expression Assay Kits currently available from Applied Biosystems. When you receive the TaqMan Protein Expression Assay Kits, Lysate Control Kits, or Core Reagents Kits, store the reagents as shown in the following tables. To order new assays that become available, see "Ordering from the Applied Biosystems website" on page 11.

Table 3 TaqMan® Protein Expression Assay Kits

Assay Kits	Contents	Rxns	Storage conditions	Part number
Human CSTB Kit (PN 4405465)	Assay Probe A, 20X	100	-15 to	4405407 (hCSTB)
Human ICAM1 Kit (PN 4405471)	(20 μL)		−25 °C	4405413 (hICAM1)
Human LIN28 Kit (PN 4405477)				4405419 (hLIN28)
Human NANOG Kit (PN 4405483)				4405425 (hNANOG)
Human OCT3/4 Kit (PN 4405489)				4405431 (hOCT3/4)
Human SOX2 Kit (PN 4405495)				4405437 (hSOX2)
	Assay Probe B, 20Χ (20 μL)			4405410 (hCSTB)
				4405416 (hICAM1)
				4405422 (hLIN28)
				4405428 (hNANOG)
				4405434 (hOCT3/4)
				4405440 (hSOX2)
	Assay Probe Dilution Buffer, 1X (0.5 mL)	100	−15 to −25 °C ‡	4405404

[‡] The Assay Probe Dilution Buffer should be kept frozen for long-term storage. However, it may be stored at 2 to 8 °C for ≤1 month after initial use.

Reagents

In addition to ordering the assay(s) for your TaqMan protein expression experiments, you should order the core reagents (Table 4) required for the experiment.

Table 4 Core Reagents Kit

Kit	Contents	Rxns	Storage conditions	Part number
TaqMan® Protein Expression Core Reagents Kit with Master	TaqMan® Protein Expression Core Reagents Base Kit (PN 4405460)	100	–15 to –25 °C	
Mix (PN 4405501)	DNA Ligase, 500Χ (10 μL)			4405389
				(DNA Ligase)
	Ligase Dilution Buffer, 1X			4405392
	(2 × 1.5 mL)			(Ligase Dilution Buffer)
	Ligation Reaction Buffer, 20X (0.7 mL)			4405395 (Ligation Reaction Buffer)
	Phosphate Buffered Saline			4412427
	(PBS), pH 7.4, 1X (1 mL)			(PBS)
	Protease, 100× (10 μL)			4405398
				(Protease)
	Universal PCR Assay, 20X			4405401
	(120 μL)			(Universal PCR Assay)
	TaqMan® Protein Expression Fast Master Mix, 2X (1.2 mL, PN 4400088)	100	2 to 8 °C‡	4400085

[‡] Optionally, the TaqMan Protein Expression Fast Master Mix can be frozen at -15 to -25 °C for long-term storage, prior to initial use.

Control cell lysate kits

Table 5 shows the TaqMan Protein Expression Lysate Control Kits currently available from Applied Biosystems. These Control Kits are for general use. You can use them as positive controls for troubleshooting. See "Ordering from the Applied Biosystems website" on page 11 for ordering instructions.

Table 5 TaqMan® Protein Expression Lysate Control Kits

Kit	Contents	Rxns [‡]	Storage conditions	Part number
Raji Kit; cells express ICAM1 and CTSB) (PN 4405448)	Lysate Control, 500 cells/µL (50 µL)	100	< –50 °C	4405380 (Raji) 4405386 (NTERA2)
NTERA2 Kit; cells express LIN28, NANOG, OCT3/4, and SOX2 (PN 4405454)	Lysate Dilution Buffer (1 mL)	100	−15 to −25 °C [§]	4405383

[‡] The number of reactions includes the reactions performed using a 2-fold dilution scheme outlined in the TaqMan®Protein Expression Assay Protocol.

[§] If necessary, the Lysate Dilution Buffer can be stored with the Lysate Control.

Other related materials

Obtain the materials shown in Table 6 for sample preparation. Unless otherwise indicated, all materials are available from major laboratory suppliers (MLS).

Table 6 Related materials

Material	Source [‡]
Centrifuge (with plate adapter)	MLS
Disposable gloves	MLS
Microcentrifuge	MLS
Pipette tips, aerosol-resistant	MLS
Pipettors (positive/air-displacement or multichannel)	MLS
Polypropylene tubes (various sizes)	MLS
Vortexer	MLS

[‡] For the MSDS of any chemical not distributed by Applied Biosystems, contact the chemical manufacturer. Before handling any chemicals, refer to the MSDS provided by the manufacturer, and observe all relevant precautions.

Ordering from the Applied Biosystems website

To search or order TaqMan Protein Expression Assays and Control Cell Lysates from the Applied Biosystems website:

- **1.** Log in to the web site, then open the TaqMan® Protein Expression Assays page:
 - a. Go to www.appliedbiosystems.com
 - b. In the Home page of the Applied Biosystems web site, click Store Log
 In (or click Log In), then log in using your customer account
 information.
 - c. After logging in, select Products ➤ Gene Expression ➤ TaqMan® Protein Expression Assays.
- **2.** In the TaqMan[®] Protein Expression Assays page, select the assay or control of interest to view details about the assay or control.
- 3. Click Add to Basket.
- **4.** Order the assay as instructed by the web site.

Appendix A Ordering Information Ordering from the Applied Biosystems website

Good Laboratory Practices

Recommended laboratory practices and guidelines

Good laboratory practices

When preparing the samples for amplification:

- Wear clean gloves and a clean lab coat (not previously worn while handling the lysate products or the reagents used during sample preparation).
- Change gloves whenever you suspect that they are contaminated.
- Maintain separate areas and dedicated equipment and supplies for:
 - Sample preparation
 - PCR setup
 - PCR amplification
 - Analysis of PCR products
- Never bring amplified PCR products into the PCR setup area.
- Open and close all sample tubes carefully. Try not to splash or spray PCR samples.
- Keep reactions and components capped as much as possible.
- Use a positive-displacement pipette or aerosol-resistant pipette tips.
- Clean lab benches and equipment periodically with 10% bleach solution.

Guidelines for sample preparation

Precise volume delivery during sample preparation is important for the performance and reproducibility of TaqMan® Protein Expression experiments. Follow the guidelines below:

- Use repeat- and/or multi-channel pipettors and ensure that all tips are properly seated prior to fluid transfer.
- Pre-aliquot reagents into a separate 96-well reaction plate for loading the multi-channel pipettor.
- Avoid creating bubbles when pipetting fluids.

Appendix B Good Laboratory Practices Recommended laboratory practices and guidelines

Appendix C Safety

This appendix covers:

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General chemical safety

Chemical hazard warning



WARNING! CHEMICAL HAZARD. Before handling any chemicals, refer to the Material Safety Data Sheet (MSDS) provided by the manufacturer, and observe all relevant precautions.



WARNING! CHEMICAL HAZARD. All chemicals in the instrument, including liquid in the lines, are potentially hazardous. Always determine what chemicals have been used in the instrument before changing reagents or instrument components. Wear appropriate eyewear, protective clothing, and gloves when working on the instrument.



WARNING! CHEMICAL HAZARD. 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. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.



WARNING! CHEMICAL STORAGE HAZARD. Never collect or store waste in a glass container because of the risk of breaking or shattering. Reagent and waste bottles can crack and leak. Each waste bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.

Chemical safety guidelines

To minimize the hazards of chemicals:

- Read and understand the Material Safety Data Sheets (MSDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. (See "About MSDSs" on page 17.)
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing). For additional safety guidelines, consult the MSDS.
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood). For additional safety guidelines, consult the MSDS.
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer's cleanup procedures as recommended in the MSDS.
- Comply with all local, state/provincial, or national laws and regulations related to chemical storage, handling, and disposal.

MSDSs

About MSDSs

Chemical manufacturers supply current Material Safety Data Sheets (MSDSs) with shipments of hazardous chemicals to new customers. They also provide MSDSs with the first shipment of a hazardous chemical to a customer after an MSDS has been updated. MSDSs provide the safety information you need to store, handle, transport, and dispose of the chemicals safely.

Each time you receive a new MSDS packaged with a hazardous chemical, be sure to replace the appropriate MSDS in your files.

Obtaining MSDSs

The MSDS for any chemical supplied by Applied Biosystems is available to you free 24 hours a day. To obtain MSDSs:

- 1. Go to www.appliedbiosystems.com, click Support, then select MSDS.
- **2.** In the Keyword Search field, enter the chemical name, product name, MSDS part number, or other information that appears in the MSDS 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 MSDSs of chemicals not distributed by Applied Biosystems, contact the chemical manufacturer.

Chemical waste safety

Chemical waste hazards



CAUTION! HAZARDOUS WASTE. Refer to Material Safety Data Sheets and local regulations for handling and disposal.



WARNING! CHEMICAL WASTE HAZARD. Wastes produced by Applied Biosystems instruments are potentially hazardous and can cause injury, illness, or death.



WARNING! CHEMICAL STORAGE HAZARD. Never collect or store waste in a glass container because of the risk of breaking or shattering. Reagent and waste bottles can crack and leak. Each waste bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.

Chemical waste safety guidelines

To minimize the hazards of chemical waste:

- Read and understand the Material Safety Data Sheets (MSDSs) provided by the manufacturers of the chemicals in the waste container before you store, handle, or dispose of chemical waste.
- Provide 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.)
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing). For additional safety guidelines, consult the MSDS.
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood). For additional safety guidelines, consult the MSDS.
- Handle chemical wastes in a fume hood.
- After emptying a waste container, seal it with the cap provided.
- Dispose of the contents of the waste tray and waste bottle in accordance with good laboratory practices and local, state/provincial, or national environmental and health regulations.

Waste disposal

If potentially hazardous waste is generated when you operate the instrument, you must:

- Characterize (by analysis if necessary) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure the health and safety of all personnel in your laboratory.

 Ensure that the instrument 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.

Biological hazard safety

General biohazard



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:

- U.S. Department of Health and Human Services guidelines published in Biosafety in Microbiological and Biomedical Laboratories (stock no. 017-040-00547-4; bmbl.od.nih.gov)
- Occupational Safety and Health Standards, Bloodborne Pathogens (29 CFR§1910.1030; 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

Chemical alerts

General alerts for all chemicals

CHEMICAL HAZARD. Avoid contact with eyes and skin. Read the MSDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

Specific chemical alerts



WARNING! 2× Cell Lysis Reagent causes eye irritation. May cause skin and respiratory tract irritation. Avoid breathing vapor. Avoid contact with eyes and skin. Keep container tightly closed. Use only with adequate ventilation. Wash thoroughly after handling. Wear appropriate protective eyewear, clothing, and gloves. This product contains sodium azide at a concentration that is considered not hazardous according to OSHA regulations. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides.



WARNING! 1× Cell Resuspension Buffer contains sodium azide at a concentration that is considered not hazardous according to OSHA regulations. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides.



WARNING! 1× pH7.4 Phosphate Buffered Saline. Read the MSDS.



WARNING! 100× Phosphatase Inhibitor Cocktail Set II. Read the MSDS.



WARNING! 100× Protease Inhibitor Cocktail Set I. Read the MSDS.

European Warnings

English	R36	Irritating to eyes.
English	R37	Irritating to respiratory system.
English	R38	Irritating to skin.
English	S24	Avoid contact with skin.
English	S26	In case of contact with eyes, rinse immediately with plenty of water and seek medical advice
English	S37	Wear suitable gloves.
English	S60	This material and/or its container must be disposed of as hazardous waste.
German	R36	Reizt die Augen.
German	R37	Reizt die Atmungsorgane.
German	R38	Reizt die Haut.
German	S24	Berührung mit der Haut vermeiden.

German	S26	Bei Berührung mit den Augen sofort gründlich mit Wasser abspülen und Arzt konsultieren.
German	S37	Geeignete Schutzhandschuhe tragen.
German	S60	Dieser Stoff und/oder sein Behälter sind als gefährlicher Abfall zu entsorgen.
French	R36	Irritant pour les yeux.
French	R37	Irritant pour les voies respiratoires.
French	R38	Irritant pour la peau.
French	S24	Eviter le contact avec la peau.
French	S26	En cas de contact avec les yeux, laver immédiatement et abondamment avec de l'eau et consulter un spécialiste
French	S37	Porter des gants appropriés.
French	S60	Eliminer le produit et/ou son récipient comme un déchet dangereux.
	_	
Spanish	R36	Irrita los ojos.
Spanish	R37	Irrita las vías respiratorias.
Spanish	R38	Irrita la piel.
Spanish	S24	Evítese el contacto con la piel.
Spanish	S26	En caso de contacto con los ojos, lávense inmediata y abundantemente con agua y acúdase a un médico
Spanish	S37	Úsese guantes adecuados.
Spanish	S60	Elimínense el producto y o recipiente como residuos peligrosos.
Italian	R36	Irritante per gli occhi.
Italian	R37	Irritante per le vie respiratorie.
Italian	R38	Irritante per la pelle.
Italian	S24	Evitare il contatto con la pelle.
Italian	S26	In caso di contatto con gli occhi,lavare immediatamente e abbondantemente con acqua e consultare un medico
Italian	S37	Usare guanti adatti.
Italian	S60	Questo materiale e/o il suo contenitore devono essere smaltiti come rifiuti pericolosi
Dutch	R36	Irriterend voor de ogen.
Dutch	R37	Irriterend voor de ademhalingswegen.
Dutch	R38	Irriterend voor de huid.
Dutch	S24	Aanraking met de huid vermijden.

Dutch	S26	Bij aanraking met de ogen onmiddellijk met overvloedig water afspoelen en deskundig medisch advies inwinnen
Dutch	S37	Draag geschikte handschoenen.
Dutch	S60	Deze stof en/of de verpakking als gevaarlijk afval afvoeren.
Swedish	R36	Irriterar ögonen.
Swedish	R37	Irriterar andningsorganen.
Swedish	R38	Irriterar huden.
Swedish	S24	Undvik kontakt med huden.
Swedish	S26	Vid kontakt med ögonen spola genast med mycket vatten och kontakta läkare.
Swedish	S37	Använd lämpliga skyddskläder.
Swedish	S60	För bort detta ämne och/eller förpackningen som farligt avfall.
Finnish	R36	Ärsyttää silmiä.
Finnish	R37	Ärsyttää hengityselimiä.
Finnish	R38	Ärsyttää ihoa.
Finnish	S24	Varottava aineen joutumista iholle.
Finnish	S26	Aineen jouduttua silmiin huuhdeltava välittömästi runsaalla vedellä ja mentävä lääkäriin
Finnish	S37	Käytettävä sopivia suojakäsineitä.
Finnish	S60	Tämä aine ja/tai sen pakkaus hävitettävä vaarallisena jätteenä.
Danish	R36	Irriterer øjnene.
Danish	R37	Irriterer åndedrætsorganerne.
Danish	R38	Irriterer huden.
Danish	S24	Undgå kontakt med huden.
Danish	S26	Kommer stoffet i øjnene, skylles straks grundigt med vand og læge kontaktes
Danish	S37	Brug egnede beskyttelseshandsker under arbejdet.
Danish	S60	Dette materiale og dets beholder skal bortskaffes som farligt affald.

Appendix C Safety Chemical alerts

Related documentation

Refer to these documents for information about Applied Biosystems PCR systems as well as protein expression reagents and kits. For additional documentation, see "How to obtain support" on page 26.

Real-time PCR system	Document	PN/SN
All	Real-Time PCR Systems Protein Expression Assays Chemistry Guide	4405780
	TaqMan® Protein Expression Assay Protocol	4405786
	Protein Expression Sample Preparation Protocol	4405785
	TaqMan [®] Protein Expression Assay Quick Reference Card	4405784
	Protein Expression Sample Preparation Quick Reference Card	4405783
7900HT Fast system Fast or standard sample blocks	Applied Biosystems 7900HT Fast Real-Time PCR System Quick Reference Card: Performing Fast Gene Quantification	4351892
	Applied Biosystems 7900HT Fast Real-Time PCR System Relative Quantitation Using Comparative C_T Getting Started Guide	4364016
	Applied Biosystems 7900HT Fast Real-Time PCR System User Bulletin: Performing Fast Gene Quantification	4369584
7300, 7500, and 7500 Fast systems	Applied Biosystems 7300/7500/7500 Fast Real-Time PCR System Relative Quantification Getting Started Guide	4347828
StepOne [™] and StepOnePlus [™] systems	Applied Biosystems StepOne [™] and StepOnePlus [™] Real-Time PCR Systems Reagent Guide	4379704
	Applied Biosystems StepOne [™] and StepOnePlus [™] Real-Time PCR Systems Relative Standard Curve and Comparative C _T Experiments Getting Started Guide	4376785

Portable document format (PDF) versions of this guide and the documents listed above are also available at **www.appliedbiosystems.com**.

Note: To open the documentation available from the Applied Biosystems web site, use the Adobe Acrobat Reader software available from **www.adobe.com**

How to obtain support

For the latest services and support information for all locations, go to **www.appliedbiosystems.com**.

At the Applied Biosystems web site, you can:

- Access worldwide telephone and fax numbers to contact Applied Biosystems Technical Support and Sales facilities.
- Search through frequently asked questions (FAQs)
- Submit a question directly to Technical Support
- Order Applied Biosystems user documents, MSDSs, certificates of analysis, and other related documents
- · Download PDF documents
- Obtain information about customer training
- Download software updates and patches

Send us your comments

Applied Biosystems welcomes your comments and suggestions for improving its user documents. You can e-mail your comments to:

techpubs@appliedbiosystems.com

IMPORTANT! The e-mail address above is only for submitting comments and suggestions relating to documentation. To order documents, download PDF files, or for help with a technical question, see "How to obtain support" on page 26.

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