USER GUIDE

applied biosystems

TaqMan® Assays for Food and Environmental Testing

Real-time PCR detection of pathogens in food and environmental samples

Publication Number MAN0009391

Revision C.0

For testing of Food and Environmental samples only.

ThermoFisher SCIENTIFIC

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About this guide

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

Revision history

Revision	Date	Description
C.0	May 2015	• Corrected dyes for TaqMan® *Salmonella* spp. & *enteritidis* Multiplex Assay Beads. • Added catalog no. for liquid format of TaqMan® *Cronobacter sakazakii* assay.
B.0	September 2014	• Updated ordering and detector dyes information. • Updated recommended DNA isolation methods. • Removed recommendation for user-supplied positive control.
A.0	October 2013	New document

1 Product information

Product description

TaqMan® Assays for Food and Environmental Testing provide a simple, reliable, and rapid method for the detection of contaminants in food and environmental samples. The assays use the polymerase chain reaction (PCR) to amplify unique microorganism-specific DNA target sequences and TaqMan® probes to detect the amplified sequences.

Note: We recommend that the user perform validation with their unique sample matrices/types to determine appropriate analysis settings (ISO 22174, 2005). Thermo Fisher Scientific offers fee-based method validation and verification services; contact **foodsafety@lifetech.com** for more information.

The assays are available in two formats:

- **Liquid assays** consist of 10X Assay Mix optimized for use with the included 2X Environmental Master Mix 2.0.
- **Lyophilized assay beads** contain all the components necessary for the real-time PCR reaction.

Both assay formats include an internal positive control (IPC) to monitor for PCR inhibition unless otherwise indicated; refer to "Ordering information and detector dyes" on page 19. The IPC also demonstrates whether or not PCR reagents are working and amplifying properly. This IPC eliminates the need for inclusion of a positive control reaction, thus reducing the risk of cross-contamination of unknown samples with the positive control.

This guide provides instructions for use on the Applied Biosystems™ 7500 Fast Real-Time PCR System. For use on other real-time PCR instruments, consult your instrument user guide.

Kit contents and storage

See "Ordering information and detector dyes" on page 19 for available assays, detector dyes, and ordering information.

Table 1 Liquid assays (100 reactions)

Components	Amount	Storage[1]
10X Assay Mix	0.30 mL	–15°C to –25°C; protect from light[2].
2X Environmental Master Mix 2.0	2 x 0.75 mL	Upon receipt: –15°C to –25°C; protect from light. After first use: 2°C to 8°C; protect from light.

[1] Refer to product label for expiration date.
[2] Excessive exposure to light may affect the fluorescent probes.

Table 2 Lyophilized assay beads (96 reactions)

Components	Amount	Storage[1]
Assay Beads, 8-tube strips	12 strips (96 tubes) 1 rack	2°C to 8°C; protect from light and moisture[2].
MicroAmp™ Optical 8-Cap Strips	12 strips (96 caps)	Room temperature.

[1] Refer to product label for expiration date.
[2] Excessive exposure to light may affect the fluorescent probes. To protect the beads from moisture, do not remove the desiccant from the pouch, and seal the pouch tightly each time you remove assay bead strips.

Required materials not included with the kits

Unless otherwise indicated, all materials are available through the Thermo Fisher Microbiology ordering process or through **www.lifetechnologies.com**. MLS: Fisher Scientific (**www.fisherscientific.com**) or other major laboratory supplier.

Item	Source
Instrument: Applied Biosystems™ 7500 Fast Real-Time PCR System	Contact your local sales representative.
Equipment	
7500 Fast Precision Plate Holder for MicroAmp™ Tube Strips	Cat. no. 4403809
MicroAmp™ 96-Well Base	Cat. no. N8010531
MicroAmp™ Cap Installing Tool	Cat. no. 4330015
MicroAmp™ Multi-removal Tool	Cat. no. 4313950

Item	Source
Plate centrifuge	MLS
Ice bucket	MLS
Vortexer	MLS
Pipettors: • Positive-displacement • Air-displacement • Multichannel	MLS
Consumables	
Aerosol-resistant pipette tips	MLS
Disposable gloves	MLS
MicroAmp™ Fast 8-Tube Strip, 0.1-mL	Cat. no. 4358293
MicroAmp™ Optical 8-Cap Strip, 300 strips	Cat. no. 4323032
Reagents	
Nuclease-free Water	Cat. no. AM9938

Recommended DNA isolation methods

Nucleic acid isolation workflow	Kit
Automated, magnetic bead-based	PrepSEQ™ Nucleic Acid Extraction Kit for Food and Environmental Testing (Cat. nos. 4480466, 4428176)
Spin columns	PrepSEQ™ Rapid Spin Sample Preparation Kit with Proteinase K (Cat. no. 4426714)
	PrepSEQ™ Rapid Spin Sample Preparation Kit – Extra Clean with Proteinase K (Cat. no. 4426715)

Workflow

Isolate DNA

▼

Perform PCR (liquid assays or lyophilized assay beads)

Create a run file

▼

Set up the PCR reactions

▼

Load and run the reactions

▼

Analyze results

Operational conditions

The Applied Biosystems™ 7500 Fast Real-Time PCR Instrument is for indoor use only and for altitudes not exceeding 2,000 m (6,500 feet) above sea level.

Table 3 Temperature and humidity requirements

Condition	Acceptable range
Temperature	15°C to 20°C Maximum change of less than 15°C per 24 hours
Humidity	20—80% relative humidity, noncondensing

2 Liquid assay PCR procedure

Create a run file

The following instructions apply to the Applied Biosystems™ 7500 Fast Real-Time PCR Instrument. For detailed instructions on setup and programming the instrument, refer to the guide accompanying your instrument or to the *7300/7500/7500 Fast Real-Time PCR System Absolute Quantitation Using Standard Curve Getting Started Guide* (Pub. no. 4347825).

1. Select **Standard Curve (Absolute Quantification)** from the Assay drop-down list.
2. Create or select the dye detectors for your assay with the Quencher Dye set to **None** or **Non-Fluorescent**.

   Refer to "Ordering information and detector dyes" on page 19.
3. Associate dyes with each reaction.
4. Name each reaction as desired.
5. Set thermal cycling conditions for the 7500 Fast Real-Time PCR Instrument according to the following table.

Stage	Stage 1 (Enzyme activation)	Stage 2 (PCR)	
Rep.	1 (Hold)	45 cycles	
		Denature	Anneal/extend
Temp.	95°C	95°C	60°C
Time	10 min	15 sec	45 sec

6. Set Sample Volume to 30 µL.
7. Select **Standard** Run Mode.

Set up the PCR reactions

1. Assemble PCR reactions in MicroAmp™ Fast 8-Tube Strips as described in the following table.

Component	Sample type	
	Test samples	Negative control
10X Assay Mix	3 µL	3 µL
2X Environmental Master Mix 2.0	15 µL	15 µL
Sample DNA	Up to 12 µL	—
Nuclease-free Water	To 30 µL total	12 µL

2. Completely seal the tubes with transparent MicroAmp™ Optical 8-Cap Strips.

   IMPORTANT! Do not use colored caps or tubes as they are not compatible with real-time PCR.

3. Mix by vortexing for 5 seconds at high speed.

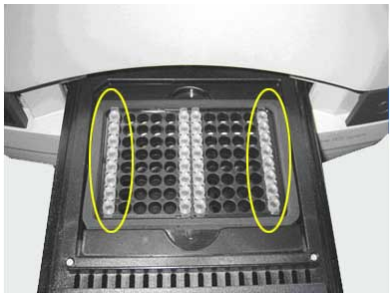
4. Centrifuge at 200 x *g* for 20 seconds to collect the contents at the bottom of the tubes.

Load and run the reactions

Use the 7500 Fast Precision Plate Holder for MicroAmp™ Tube Strips in the instrument.

1. Open the instrument loading block and place the prepared tube strips in a vertical position in the center of the block of the 7500 Fast Real-Time PCR Instrument.

   If columns 1 and 12 are not used, fill them with empty strips. This will balance the block to avoid damage of tubes.



2. Open the run file created in “Create a run file“ on page 10.

3. Close the instrument loading block and start the run.

Analyze results

The general process for analyzing results from TaqMan® Assays for Food and Environmental Testing is:

1. View the amplification plots for all reactions to make sure that they appear normal.
2. Set the baseline and threshold values.
3. Use the relative standard curve or the comparative C_T method to analyze your data.

The details of data analysis depend on the real-time PCR instrument that you use; refer to the appropriate user guide for instructions on how to analyze your data.

3 Lyophilized assay beads PCR procedure

Create a run file

The following instructions apply to the Applied Biosystems™ 7500 Fast Real-Time PCR Instrument. For detailed instructions on setup and programming the instrument, refer to the guide accompanying your instrument or to the *7300/7500/7500 Fast Real-Time PCR System Absolute Quantitation Using Standard Curve Getting Started Guide* (Pub. no. 4347825).

1. Select **Standard Curve (Absolute Quantification)** from the Assay drop-down list.
2. Create or select the dye detectors for your assay with the Quencher Dye set to **None** or **Non-Fluorescent**.
   Refer to "Ordering information and detector dyes" on page 19.
3. Associate dyes with each reaction.
4. Name each reaction as desired.
5. Set thermal cycling conditions for the 7500 Fast Real-Time PCR Instrument according to the following table.

Stage	Stage 1 (Enzyme activation)	Stage 2 (PCR)	
Rep.	1 (Hold)	40 cycles	
		Denature	Anneal/extend
Temp.	95°C	95°C	60°C
Time	2 min	3 sec	30 sec

6. Set Sample Volume to 30 µL.
7. Select **Fast** Run Mode.

Set up the PCR reactions

Each reaction requires 30 µL of sample.

1. Place an appropriate number of 8-tube strips containing assay beads in a 96-well base, based on the number of samples and controls that you plan to run, and label appropriately.

   If needed, gently tap the tubes to move the assay beads to the bottom of all tubes.

   For 8-tube strips with seven or fewer reactions, add additional empty tubes as needed so that each strip contains a full set of 8 tubes.

2. Carefully remove the caps from the 8-tube strips and discard the caps.

3. Add up to 30 µL of sample or control to each assay bead.

   Dispense all unknown samples first, followed by negative control(s).

   Use a new pipette tip for each different sample.

4. Completely seal the tubes with the transparent MicroAmp™ Optical 8-Cap Strips provided in the kit.

   IMPORTANT! Do not use colored caps or tubes as they are not compatible with real-time PCR.

5. Mix by vortexing for 5 seconds at high speed.

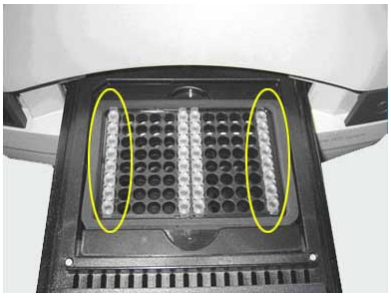
6. Centrifuge at 200 x *g* for 20 seconds to collect the contents at the bottom of the tubes.

Load and run the reactions

Use the 7500 Fast Precision Plate Holder for MicroAmp™ Tube Strips in the instrument.

1. Open the instrument loading block and place the prepared tube strips in a vertical position in the center of the block of the 7500 Fast Real-Time PCR Instrument.

   If columns 1 and 12 are not used, fill them with empty strips. This will balance the block to avoid damage of tubes.



2. Open the run file created in "Create a run file" on page 13.
3. Close the instrument loading block and start the run.

Analyze results

The general process for analyzing results from TaqMan® Assays for Food and Environmental Testing is:

1. View the amplification plots for all reactions to make sure that they appear normal.
2. Set the baseline and threshold values.
3. Use the relative standard curve or the comparative C_T method to analyze your data.

The details of data analysis depend on the real-time PCR instrument that you use; refer to the appropriate user guide for instructions on how to analyze your data.

Troubleshooting

Observation	Possible cause	Recommended action
In negative control wells, no IPC signal is detected.	Carryover contamination from samples containing a high copy number of target DNA, resulting in preferential amplification of the target-specific DNA in the negative control wells.	To correct carryover contamination, repeat the assay using fresh aliquots of all reagents and clean pipetting equipment.
	A problem occurred with IPC amplification.	To determine whether IPC amplification is a problem, examine unknown wells for an IPC signal. If an IPC signal is present, IPC amplification is not a problem.
In negative control wells, target-specific signal is detected.	Carryover contamination occurred.	1. Repeat the assay using fresh aliquots of all reagents and clean pipetting equipment. 2. If the negative control continues to show contamination, repeat the assay using a new kit. 3. If the negative control continues to show contamination, contact Technical Support.
In unknown wells, no IPC or target-specific signal is detected.	Inhibition of PCR occurred.	Dilute the sample 1:5 with Nuclease-free Water to dilute PCR inhibitors, and repeat the assay. If PCR remains inhibited, repeat the sample preparation. Refer to other troubleshooting suggestions for removal of Magnetic Particles or particulate residue from the DNA sample.
In unknown sample wells, no IPC is detected, but target specific signal (C_T <35) is detected.	A high copy number of target DNA exists in samples, resulting in preferential amplification of the target-specific DNA.	No action is required. The result may be considered positive. For some assays, both FAM™ and VIC™ targets must amplify. If only one amplifies, but the IPC does not, the result is considered inconclusive.

Observation	Possible cause	Recommended action
Multicomponent plot signals for FAM™, VIC™, NED™, and LIZ™ dyes increase/decrease during cycles 1–15, but the overall curve and result are not affected.	Incomplete mixing and dissolution of the lyophilized bead with sample or control.	After addition of 30 µL of sample or no template control to the beads and capping the tubes: 1. Vortex strips at high speed for about 10 seconds, and centrifuge the strips at 200–600 × *g* for about 10 seconds. 2. Vortex the strips again on high speed for about 10 seconds, and centrifuge the strips at 200–600 × *g* for about 1 minute. Ensure that all liquid is at the bottom of the tubes and the beads are fully dissolved before proceeding.
Amplicon contamination.	• Contamination was introduced into the PCR clean area from post-amplification reaction tubes that were either opened in the clean area or brought into the PCR clean area from contaminated gloves or solutions. • Contamination was introduced into the real-time PCR instrument from crushed and broken PCR reaction tubes.	Prepare negative control samples using at least one 8-tube strip of Assay Beads. 1. Divide the assay beads into two sets. a. To the first set of assay beads, add 30 µL of Nuclease-free Water. b. To the second set of assay beads, add 29 µL of Nuclease-free Water plus 1 µL of 1 U/µL Uracil DNA Glycosylase (Cat. no. 18054-015). 2. Run samples on the 7500 Fast Real-Time PCR Instrument using SDS software and select **Fast 7500** run mode. 3. Under the instrument tab: • Select **Add Step** to stage 1 of the PCR cycle that consists of 10 minutes at 50°C. • Extend the 95°C step from 20 seconds to 10 minutes. Amplicon contamination is indicated by target-specific signal in the –UNG samples and no target-specific signal in +UNG samples. If the instrument block was contaminated, consult the *7300/7500/7500 Fast Real-Time PCR System Absolute Quantitation Using Standard Curve Getting Started Guide* (Pub. no. 4347825) and/or contact a service representative to clean the instrument.
Small ΔRn.	PCR efficiency is poor.	Recheck the concentration of the reagents.
	Quantity of starting target is low (low copy number of target).	Increase the quantity of the starting target.

Observation	Possible cause	Recommended action
High standard deviation of replicates (inconsistent data, C_T varies).	Inefficient mixing of reagents.	Increase the length of time that you mix the reagents.
		Validate your mixing process by running a replicate plate.
	Pipetting was inaccurate.	Check the calibration of the pipettes.
		Pipette more than 5 µL of sample.
	Threshold was set improperly.	Set the threshold above the noise and where the replicates are tightest. Refer to your real-time PCR system user documentation for procedures on setting the threshold.
	Low concentration of target.	Rerun the assay using more template.

B

Supplemental information

Ordering information and detector dyes

Visit **www.lifetechnologies.com/foodsafety** for an up-to-date of list of available TaqMan® Food and Environmental Assays, or contact **foodsafety@lifetech.com** or your regional Food Safety sales representative.

If you cannot find a predesigned assay to suit your needs, contact **foodsafety@lifetech.com** or your regional Food Safety sales representative.

Assay name	Detector dye				Catalog no.	
	FAM™	VIC™	NED™	LIZ™	Liquid	Lyophilized
Meat ID kits						
TaqMan® Beef Species ID	Beef	IPC	N/A	N/A	4484972	4485024
TaqMan® Chicken Species ID	Chicken	IPC	N/A	N/A	4484973	4485025
TaqMan® Porcine Species ID	Pork	IPC	N/A	N/A	4484974	4485026
ISO STEC design						
TaqMan® EAE, ISO	EAE	IPC	N/A	N/A	4485004	4485060
TaqMan® STX1, ISO	STX1	IPC	N/A	N/A	4485005	4485061
TaqMan® STX2, ISO	STX2 (w/o STX2F)	IPC	N/A	N/A	4485006	4485062
TaqMan® STEC Screening, ISO[1]	O157	STX1/STX2	IPC	EAE	—	4485075
TaqMan® STEC O45 & O121, ISO[1]	O45	IPC	O121	N/A	—	4485082
TaqMan® STEC O26, O103 & O145, ISO[1]	O26	O103	O145	IPC	—	4485083
TaqMan® STEC O111 & O104, ISO[1]	O104	O111	IPC	N/A	—	4485084
EPA beach water assays						
TaqMan® *Bacteroides* spp.	*Bacteroides*	IPC	N/A	N/A	4484990	4485044
TaqMan® *Enterococcus* spp.	*Enterococcus*	IPC	N/A	N/A	4484991	4485045
TaqMan® *Oncorhynchus keta*	*O. keta*	IPC	N/A	N/A	4484992	4485046

Assay name	Detector dye				Catalog no.	
	FAM™	VIC™	NED™	LIZ™	Liquid	Lyophilized
MLG STEC						
TaqMan® STEC O103 & O145, MLG[2]	O145	O103	N/A	16S rRNA[3]	4485007	4485063
TaqMan® STEC O26 & O111, MLG[2]	O26	O111	N/A	16S rRNA[3]	4485008	4485064
TaqMan® STEC O45 & O121, MLG[2]	O45	O121	N/A	16S rRNA[3]	4485009	4485065
TaqMan® STEC STX & EAE, MLG[2]	STX1 & STX2 (except STX2F)	EAE	N/A	16S rRNA[3]	4485010	4485066
Assays by target pathogen						
Cronobacter						
TaqMan® *Cronobacter sakazakii*	*C. sakazakii*	IPC	N/A	N/A	4382492	4485034
TaqMan® *Cronobacter* spp.	*Cronobacter*	IPC	N/A	N/A	4484981	4485035
Campylobacter						
TaqMan® *Campylobacter* Multiplex	*C. coli*	*C. jejuni*	IPC	*C. lari*	4484975	4485027
TaqMan® *Campylobacter coli*	*C. coli*	IPC	N/A	N/A	4484976	4485028
TaqMan® *Campylobacter lari*	*C. lari*	IPC	N/A	N/A	4484979	4485032
Escherichia coli						
TaqMan® *Escherichia coli* O104	O104	IPC	N/A	N/A	4484982	4485036
TaqMan® *Escherichia coli* O104:H4	O104:H4	IPC	N/A	N/A	4484983	4485037
TaqMan® *Escherichia coli* 2011 O104:H4	2011 O104:H4	IPC	N/A	N/A	4484984	4485038
TaqMan® *Escherichia coli* spp.	*E. coli* spp.	IPC	N/A	N/A	4484985	4485039
TaqMan® Shiga-like Toxin (STX1)	STX1	IPC	N/A	N/A	4485001	4485057
TaqMan® Shiga-like Toxin (STX2)	STX2	IPC	N/A	N/A	4485002	4485058
TaqMan® Shiga-like Toxin (STX1/ STX2) Multiplex	STX2	STX1	IPC	N/A	4485003	4485059
TaqMan® Verotoxin-producing *Escherichia coli* VT1	VT1	IPC	N/A	N/A	4485016	4485072
TaqMan® Verotoxin-producing *Escherichia coli* VT2	VT2	IPC	N/A	N/A	4485017	4485073

Assay name	Detector dye				Catalog no.	
	FAM™	VIC™	NED™	LIZ™	Liquid	Lyophilized
TaqMan® Verotoxin-producing *Escherichia coli* VT1/VT2 Multiplex	VT2	VT1	IPC	N/A	4485018	4485074
Pseudomonas						
TaqMan® *Pseudomonas aeruginosa*	*P. aeruginosa*	IPC	N/A	N/A	—	4485047
Salmonella						
TaqMan® *Salmonella* spp. & *enteritidis* Multiplex Assay Beads	*S. enteritidis*	*Salmonella* spp.	IPC	N/A	—	4485086
TaqMan® *Salmonella* Newport	*S.* Newport	IPC	N/A	N/A	4484993	4485048
TaqMan® *Salmonella* spp. Ultimate	*Salmonella*[4]		IPC	N/A	4484994	4485049
TaqMan® *Salmonella* Heidelberg	*S.* Heidelberg[4]		IPC	N/A	4484995	4485050
TaqMan® *Salmonella* Hadar	*S.* Hadar[4]		IPC	N/A	4484996	4485051
TaqMan® *Salmonella* Senftenberg	*S.* Senftenberg	IPC	N/A	N/A	4484997	4485052
TaqMan® *Salmonella* Typhimurium	*S.* Typhimurium[4]		IPC	N/A	4484998	4485053
Staphylococcus						
TaqMan® *Staphylococcus aureus*	*S. aureus*	IPC	N/A	N/A	—	4485054
Vibrio						
TaqMan® *Vibrio* Multiplex	*V. parahaemolyticus*	*V. cholerae*	IPC	*V. vulnificus*	4485012	4485068
TaqMan® *Vibrio cholera*	*V. cholerae*	IPC	N/A	N/A	4485013	4485069
TaqMan® *Vibrio parahaemolyticus*	*V. parahaemolyticus*	IPC	N/A	N/A	4485014	4485070
TaqMan® *Vibrio vulnificus*	*V. vulnificus*	IPC	N/A	N/A	4485015	4485071
Quality testing organisms						
TaqMan® *Allicyclobacillus acidoterrestris*	*A. acidoterrestris*	IPC	N/A	N/A	4484970	4485022
TaqMan® *Allicyclobacillus* Multiplex	*A. acidoterrestris*	IPC	*A. acidocaldarius*	N/A	4484971	4485023

Assay name	Detector dye				Catalog no.	
	FAM™	VIC™	NED™	LIZ™	Liquid	Lyophilized
TaqMan® *Candida albicans*	*C. albicans*	IPC	N/A	N/A	4484980	4485033
TaqMan® *Gluconacetobacter liquefaciens*	*G. liquefaciens*	IPC	N/A	N/A	4484986	4485040

[1] See ISO/TS 1316:2012(E) guidelines.
[2] See USDA MLG 5B (non-O157) guidelines.
[3] IPC is not included.
[4] Both FAM™ **AND** VIC™ must be positive for the target to be present (triplex assay).

Good laboratory practices for PCR and RT-PCR

When preparing samples for PCR or RT-PCR amplification:

- Wear clean gloves and a clean lab coat (not previously worn while handling amplified products or during sample preparation).
- Change gloves whenever you suspect that they are contaminated.
- Maintain separate areas and dedicated equipment and supplies for:
  - Sample preparation and reaction setup.
  - Amplification and analysis of products.
- Do not bring amplified products into the reaction setup area.
- Open and close all sample tubes carefully. Avoid splashing or spraying samples.
- Keep reactions and components capped as much as possible.
- Use a positive-displacement pipettor or aerosol-resistant barrier pipette tips.
- Clean lab benches and equipment periodically with 10% bleach solution or DNA*Zap*™ Solutions (Cat. no. AM9890).

C

Safety

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.

Biological hazard safety

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. All work should be conducted in properly equipped facilities using the appropriate safety equipment (for example, physical containment devices). Safety equipment also may include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 5th Edition, HHS Publication No. (CDC) 21-1112, Revised December 2009; found at:
  www.cdc.gov/biosafety/publications/bmbl5/BMBL.pdf
- World Health Organization, *Laboratory Biosafety Manual*, 3rd Edition, WHO/CDS/CSR/LYO/2004.11; found at:
  www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf

Documentation and support

Related documents

Visit the product web page at **www.lifetechnologies.com** for instrument user guides for your Thermo Fisher Scientific real-time PCR instrument.

Customer and technical support

Visit **www.lifetechnologies.com/support** for the latest in services and support, including:

- Worldwide contact telephone numbers
- Product support, including:
  - Product FAQs
  - Software, patches, and updates
- Order and web support
- Product documentation, including:
  - User guides, manuals, and protocols
  - Certificates of Analysis
  - Safety Data Sheets (SDSs; also known as MSDSs)

    Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Food Safety support

Website: **www.lifetechnologies.com/foodsafety**

Support email: **foodsafety@lifetech.com**

Phone number in North America: 1-800-500-6855

Phone number outside of North America: Visit **www.lifetechnologies.com/support**, select the link for phone support, and select the appropriate country from the dropdown menu.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at **www.lifetechnologies.com/termsandconditions**. If you have any questions, please contact Life Technologies at **www.lifetechnologies.com/support**.

References

ISO. 2005. Microbiology of food and animal feeding stuffs -- Polymerase chain reaction (PCR) for the detection of food-borne pathogens -- General requirements and definitions. Reference number 22174:2005.

U.S. Department of Agriculture, Food Safety and Inspection Service, Microbiology Laboratory Guidebook. Detection and isolation of non-O157 Shiga toxin-producing *Escherichia coli* (STEC) from meat products and carcass and environmental sponges. Microbiology Laboratory Guidebook. MLG 5B.05.

ISO. 2012. Microbiology of food and animal feed – Real-time polymerase chain reaction (PCR)-based method for the detection of food-borne pathogens – Horizontal method for the detection of Shiga toxin-producing *Escherichia coli* (STEC) and the determination of O157, O111, O26, O103 and O145 serogroups. Reference number ISO/TS 13136:2012(E).

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20 May 2015

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