USER GUIDE

applied biosystems by life technologies™

# PrepSEQ® Rapid Spin Sample Preparation Kits for Food Testing: *E. coli* O157:H7

**Catalog Numbers** 4407760, 4426714 (with Proteinase K), 4413269 (Extra Clean), 4426715 (Extra Clean with Proteinase K)

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BY AFNOR CERTIFICATION NF VALIDATION EN ISO 16140

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(End of validity: refer to certificate available at www.afnor-validation.com]

ALTERNATIVE ANALYTICAL METHODS FOR AGRIBUSINESS

Certified by AFNOR Certification

**For testing of Food and Environmental samples only.**

life technologies™

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**TRADEMARKS:**

# Contents

**IMPORTANT!** Before using this product, read and understand the information in Appendix B, "Safety" on page 21.

**WARNING!** *E. coli* O157:H7 is a Biosafety Level 2 (BSL-2) organism. Care must be taken when handling samples that may contain *E. coli* O157:H7. Laboratory personnel must be adequately trained to handle pathogens before being permitted to analyze samples for *E. coli* O157:H7. Laboratory personnel must wear appropriate protective equipment, which includes but is not limited to: protective eyewear, face shield, clothing/lab coat, and gloves. Extreme precautions should be taken with contaminated sharp items. Access to the laboratory should be limited when work is being conducted. Waste should be disposed of in compliance with local and national legislation as appropriate.

# Product description

The PrepSEQ® Rapid Spin Sample Preparation Kits provide a simple way to prepare DNA from broth cultures.

For additional background information, refer to Appendix A on page 19.

# Materials and equipment

## Kit contents

The PrepSEQ® Rapid Spin Sample Preparation Kits for Food Testing contain reagents for 100 sample preparations. Kit components and storage conditions are shown in the following tables.

**PrepSEQ® Rapid Spin Sample Preparation Kit (Cat. no. 4407760)**

| Item | Quantity or volume | Storage |
|---|---|---|
| Spin columns | 100 | Room temperature (23 ±5°C) |
| Microcentrifuge tubes, 1.5 mL | 100 | Room temperature (23 ±5°C) |
| Lysis Buffer, 1 bottle | 5 mL | 5 ±3°C |

| **PrepSEQ® Rapid Spin Sample Preparation Kit with Proteinase K (Cat. no. 4426714)** | | |
|---|---|---|
| Item | Quantity or volume | Storage |
| Spin columns | 100 | Room temperature (23 ±5°C) |
| Microcentrifuge tubes, 1.5 mL | 100 | Room temperature (23 ±5°C) |
| Lysis Buffer, 1 bottle | 5 mL | 5 ±3°C |
| Proteinase K (20 mg/mL), 1 tube | 1.25 mL | Below –18°C |

| **PrepSEQ® Rapid Spin Sample Preparation Kit – Extra Clean (Cat. no. 4413269)** | | |
|---|---|---|
| Item | Quantity or volume | Storage |
| Spin columns | 100 | Room temperature (23 ±5°C) |
| Microcentrifuge tubes, 1.5 mL | 2 × 100 | Room temperature (23 ±5°C) |
| Lysis Buffer, 1 bottle | 5 mL | 5 ±3°C |

| **PrepSEQ® Rapid Spin Sample Preparation Kit – Extra Clean with Proteinase K (Cat. no. 4426715)** | | |
|---|---|---|
| Item | Quantity or volume | Storage |
| Spin columns | 100 | Room temperature (23 ±5°C) |
| Microcentrifuge tubes, 1.5 mL | 2 × 100 | Room temperature (23 ±5°C) |
| Lysis Buffer, 1 bottle | 5 mL | 5 ±3°C |
| Proteinase K (20 mg/mL), 1 tube | 1.25 mL | Below –18°C |

**Note:** Parts may ship separately depending on configuration and storage conditions.

## Materials not included in the kit

The following table includes materials and equipment for using (but not included in) the PrepSEQ® Rapid Spin Sample Preparation Kits. Unless otherwise indicated, many of the listed items are available from major laboratory suppliers (MLS).

**Equipment, consumables, and reagents‡**

| Item | Source |
|---|---|
| **Equipment** | |
| Benchtop microcentrifuge | Eppendorf 5415D or equivalent |
| Block heater, 56°C | MLS |
| Block heater, 95°C | MLS |
| Rack for 1.5-mL tubes | MLS |
| Homogenizer, Stomacher® 400 Laboratory Blender | Seward #0400/001/AJ or equivalent |
| Vortexer | MLS |
| **Consumables** | |
| Disposable gloves | MLS |
| Micropipette tips, aerosol-resistant | MLS |
| Pipettors:<br>• Positive-displacement<br>• Air-displacement | MLS |
| Whirl-Pak® Filter Bags, 10” × 15”, 92 oz. (Stomacher® bags with mesh) | Nasco #B01488WA or equivalent |
| Whirl-Pak® Filter Bags, 6” × 9”, 24 oz., 250/pkg (Stomacher® bags with mesh) | Nasco #B01348WA or equivalent |
| Whirl-Pak® Bags, 6” × 9”, 24 oz. (Stomacher® bags without mesh) | Nasco #B01297WA or equivalent |
| **Reagents** | |
| Brain Heart Infusion (BHI) Broth | MLS |
| Buffered Peptone Water (BPW) | MLS |
| Nuclease-free water | Life Technologies Cat. no. AM9938 |

‡ The materials listed here have been validated for use with this kit. Results may vary if substituted products from other vendors are used instead.

# PrepSEQ® Rapid Spin Sample Preparation Kits: *E. coli* O157:H7



## Before you begin

Before starting your sample extraction:

- Set the block heater temperature to 95°C. If using the Proteinase K treatment, set another block heater to 56°C.

  **Note:** For some foods with high protein content, use the PrepSEQ® Rapid Spin Sample Preparation Kit – Extra Clean with Proteinase K (Cat. no. 4426715). See table on page 10 to see food types requiring proteinase K.

- Label the 1.5-mL microcentrifuge tubes.
- For processing multiple samples in workflows that require Proteinase K treatment, we recommend preparing a Proteinase K-Lysis Buffer mix:

  a. Premix 5 µL of Proteinase K (20 mg/mL) with 50 µL of Lysis Buffer for each sample (use a clean appropriately-sized container for mixing).

  b. Multiply volumes by the number of samples plus 10% for overage.

  c. Mix well to disperse Proteinase K in Lysis Buffer.

  d. Use immediately, or store on ice until ready to use.

# General differences between PrepSEQ® Rapid Spin workflows

This document presents protocols for three different workflows, with general differences outlined in the following table. To determine which enrichment protocol you should follow for your sample, consider the sample amount and select a workflow based on your preferred media and enrichment time. Kit workflows are described on page 11.

| Protocol | Sample amount | Media | Enrichment time‡ | Enrichment volume required for sample prep | Food type/ Proteinase K requirement |
|---|---|---|---|---|---|
| Enrichment workflow A | 25 g or 25 mL of food | Prewarmed Brain Heart Infusion (BHI) | 6–8 hr<br><br>(8–10 hr for juices) | 750 µL | Animal products§: with Proteinase K |
| | | | | | Non-animal products: without Proteinase K |
| Enrichment workflow B | 25 g or 25 mL of food | Buffered Peptone Water (BPW) | 16–20 hr | 750 µL | Animal products: with Proteinase K (optional)#<br><br>Non-animal products: without Proteinase K |
| Enrichment workflow C | 375 g of food | BPW | 16–20 hr | 750 µL | All foods: with Proteinase K |

**IMPORTANT!** Enrichment workflow C (sample amount = 375 g of food) was included in AOAC validation studies but not in NF VALIDATION studies.

‡ All enrichments are incubated at 42 ±1°C.
§ Animal products include ground beef, beef trim, fish, and milk.
# For Enrichment workflow B, Proteinase K was used in NF VALIDATION studies; it was not used in AOAC validation studies.

# Kit workflow

The following is a sample-preparation workflow for procedures starting on page 12.

**Enrichment A:** *E. coli* O157:H7
**6–8-hr** enrichment for 25 g or 25 mL of food sample

Step 1: Add 225 mL of prewarmed BHI to 25 g or 25 mL of food sample.

Step 2: Homogenize the food sample.

Step 3: Incubate at 42 ±1°C for 6–8 hr under static conditions (8–10 hr for juices).

**Enrichment B:** *E. coli* O157:H7
**16–20-hr** enrichment for 25 g or 25 mL of food sample

Step 1: Add 225 mL of BPW to 25 g or 25 mL of food sample.

Step 2: Homogenize the food sample.

Step 3: Incubate at 42 ±1°C for 16–20 hr under static conditions.

**Enrichment C:** *E. coli* O157:H7
**16–20-hr** enrichment for 375 g of food sample

Step 1: Add 1.5 L of BPW to 375 g of food sample.

Step 2: Homogenize the food sample.

Step 3: Incubate at 42 ±1°C for 16–20 hr under static conditions.

**Sample preparation**

Step 1: Insert a spin column into a labeled tube.

Step 2: Load 750 µL of sample onto the spin column and cap the column.

Step 3: Load the tube into the microcentrifuge.

Step 4: Microcentrifuge at 12,000–16,000 × *g* for approximately 3 min.

Step 5: Remove the tube from the microcentrifuge, then discard the used spin column.

Step 6: Aspirate, then discard the supernatant.

- **Enrichment A**: *animal products only*
- **Enrichment B**: *animal products (in NF-VALIDATION validated workflow)*
- **Enrichment C**: *all food types*

Step 7a: Add 55 µL of Proteinase K-Lysis Buffer mix to the pellet.

- **Enrichment A**: *non-animal products only*
- **Enrichment B**: *all food types (in AOAC-validated workflow)*
- **Enrichment B**: *non-animal products (in NF-VALIDATION validated workflow)*

Step 7b: Add 50 µL of Lysis Buffer to the pellet.

Step 8: Resuspend by pipetting up and down, or vortex until the pellet is well dispersed.

For samples with high lipid content (PrepSEQ® Rapid Spin Extra Clean protocol)
For all other samples, skip Step 9 and proceed directly to Step 10.

Step 9: Transfer the lysate to a clean 1.5-mL tube.

For all other samples

Step 10: For samples treated with Proteinase K-Lysis Buffer *only*, incubate at 56 ±1°C for at least 30 min.
For other samples, go directly to step 11.

Step 11: Incubate at 97 ±2°C for 12 ±2 min. Allow the sample to cool for approx. 2 min at room temperature (23 ±5°C).

Step 12: Microcentrifuge 12,000–16,000 × *g* for approximately 1 min.

Step 13: Add 250 µL of water. Mix well.

Step 14: Microcentrifuge at 12,000–16,000 × *g* for approximately 1 min.

Step 15: Proceed with PCR, or store the tube below –18°C.

# Protocol

## Enrichment of food sample

1. Add to the food sample the appropriate enrichment media depending upon the *E. coli* O157:H7 workflow selected from the following options:
   - **Enrichment workflow A** – For 6–8-hour enrichment of 25 g or 25 mL of food (8–10-hour enrichment for juices), add 225 mL of prewarmed (42 ±1°C) Brain Heart Infusion (BHI) broth.
   - **Enrichment workflow B** – For 16–20-hour enrichment of 25 g or 25 mL of food, add 225 mL of Buffered Peptone Water (BPW).
   - **Enrichment workflow C** – For 16–20-hour enrichment of 375 g of food, add 1.5 L of BPW.

   **Note:** For 375-g food samples, use only Enrichment workflow C.

2. Homogenize the food sample:
   - For 375-g food samples (Enrichment workflow C), use a filtered 10” × 15” Stomacher® bag (Nasco #B01488WA or equivalent) and squeeze the bag 5–10 times to break up food chunks.
   - For solid foods (such as chicken wings), use a filtered Stomacher® bag and mix by hand by squeezing the bag 5–10 times.
   - For coarse food types (such as meat, poultry, and seafood), use a filtered 6” × 9” Stomacher® bag (Nasco #B01348WA or equivalent) and stomach with the speed setting set to **Norm** for 1 minute (Stomacher® 400 Laboratory Blender or equivalent).
   - For soft food types (such as mayonnaise), use a standard 6” × 9” Stomacher® bag (Nasco #B01297WA or equivalent) and stomach with the speed setting set to **Norm for 1 minute** (Stomacher® 400 Laboratory Blender or equivalent).
   - For liquids or powdered foods, use a standard 6” × 9” Stomacher® bag (Nasco #B01297WA or equivalent) and mix by hand.

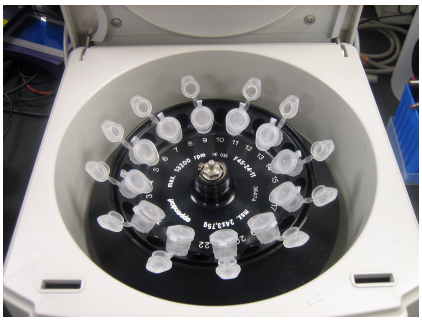
3. Incubate the food sample at 42 ±1°C under static conditions for the following amount of time:
   - **Enrichment workflow A** – 6–8 hours (8–10 hours for juices) to enable same-day result. For convenience, the sample can be enriched in BHI for up to 16 hours.
   - **Enrichment workflow B** – 16–20 hours.
   - **Enrichment workflow C** – 16–20 hours.

   **Note:** The minimum enrichment incubation time is 6 hours for Enrichment workflow A and 16 hours for Enrichment workflows B and C.

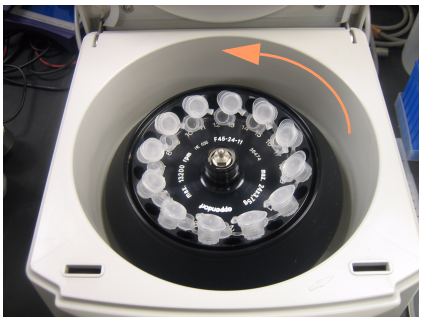
## Sample preparation

1. Insert a spin column into a labeled tube.
2. Load 750 µL of your enriched sample onto the spin column and cap the column.
3. Load the tube with its spin column into the microcentrifuge. Place the tube cap hinge toward the inside of the rotor (see the following figure labelled "Correct position of the tube cap"). Otherwise the cap hinge interferes with installation of the rotor lid.

   **Note:** To prevent damage to the tube cap during centrifugation, position the cap in the opposite direction of rotation.



Incorrect position of the tube cap



Correct position of the tube cap

4. Microcentrifuge the tube at 12,000–16,000 × *g* for approximately 3 minutes.
5. Remove the tube from the microcentrifuge, then discard the used spin column.

6. Aspirate, then discard the supernatant.

**IMPORTANT!** Remove the supernatant as completely as possible, including any excess liquid on the sides of the tube. Remove droplets by circling the inside of the tube with the pipettor and pushing into supernatant for removal by aspiration.

**IMPORTANT!** For samples that contain a fat layer following centrifugation, indicated as a distinct top layer, remove the fat layer as follows:

*For liquid fat layer* (for example, as found in milk samples):

1. Use a P1000 pipettor to remove fat from the top surface by aspirating in a circular motion without disturbing the pellet.
2. Continue to collect supernatant from the top surface until all the supernatant is removed.
3. Discard the supernatant into a waste container.

***or***

*For solid fat layer* (for example, as found in infant formula samples):

1. Use a pipettor to gently dislodge the fat layer without disturbing the pellet.
2. Pour off the supernatant and fat layer using a single quick motion.
3. Aspirate the supernatant from the top surface using a pipettor until all the supernatant is removed.
4. Discard the supernatant into a waste container.

7. Depending on the enrichment and food type, add Proteinase K-Lysis Buffer mix or Lysis Buffer (only) using one of the two following options:
   - Add 55 µL of Proteinase K-Lysis Buffer mix (see page 9) to the pellet for the following enrichments and foods:
     – **Enrichment workflow A** – For 6–8-hour enrichment of 25 g or 25 mL of foods (animal products, such as ground beef, beef trim, fish, and milk).
     – **Enrichment workflow B** – For 16–20-hour enrichment of 25 g of food product of animal origin.

     **Note:** In the context of NF validation, proteinase K was used for beef (or animal-origin samples) but not in the AOAC-validation study.

     – **Enrichment workflow C** – For 16–20-hour enrichment of 375 g of foods (all food types).

     **IMPORTANT!** Store Proteinase K-Lysis Buffer mix on ice until ready to use.

***or***

- Add 50 µL of Lysis Buffer to the pellet for the following enrichments and foods:
  - **Enrichment workflow A** – For 6–8-hour enrichment of 25 g or 25 mL of foods (8–10-hour enrichment for juices); non-animal products only.
  - **Enrichment workflow B** – For 16–20-hour enrichment of 25 g or 25 mL of foods (all food types).

8. Resuspend by pipetting up and down, or vortex until the pellet is well dispersed.

9. **Rapid Spin Extra Clean protocol *only*** – If the food sample has high lipid content, transfer the mixture into a clean 1.5-mL tube (avoid transferring the fat when transferring the mixture). The pellet must be well dispersed in the Lysis Buffer prior to transfer.

   **IMPORTANT!** During the transfer there will be residual fat on the sides of the original tube. Avoid contact with the fat and transfer only the Lysis Buffer containing the resuspended pellet into a clean tube.

   **IMPORTANT!** We recommend the Rapid Spin Extra Clean protocol step for food samples with high lipid content [such as infant formula, whole milk, smoked salmon (lox), and chicken wing samples]. This protocol is also recommended with some 375-g food samples depending on the sample's fat content.

   **Note:** Inhibition of PCR with the detection assay will be indicated by warning calls with the RapidFinder™ Express software or non-detection of the internal positive control with SDS or StepOne® software. If your matrix has high fat content and if inhibition of PCR is frequently observed, we recommend applying the Rapid Spin Extra Clean protocol step to the matrix.

   ***or***

   For all other samples, proceed to step 10.

10. For samples using Proteinase K-Lysis Buffer *only*:

    Cap the tube, then incubate at 56 ±1°C for at least 30 minutes to activate the Proteinase K. Proceed to step 11.

    ***or***

    For samples using Lysis Buffer, proceed to step 11.

11. For all samples, incubate at 97 ±2°C for 12 ±2 minutes to lyse samples, then allow the sample to cool for approximately 2 minutes at room temperature (23 ±5°C).

12. Microcentrifuge the tube at 12,000–16,000 × *g* for approximately 1 minute to bring down condensation.

13. Add 250 µL of water. Mix well.

**IMPORTANT!** Use PCR-clean water (Cat. no. AM9938). Autoclaved water should not be considered PCR-clean.

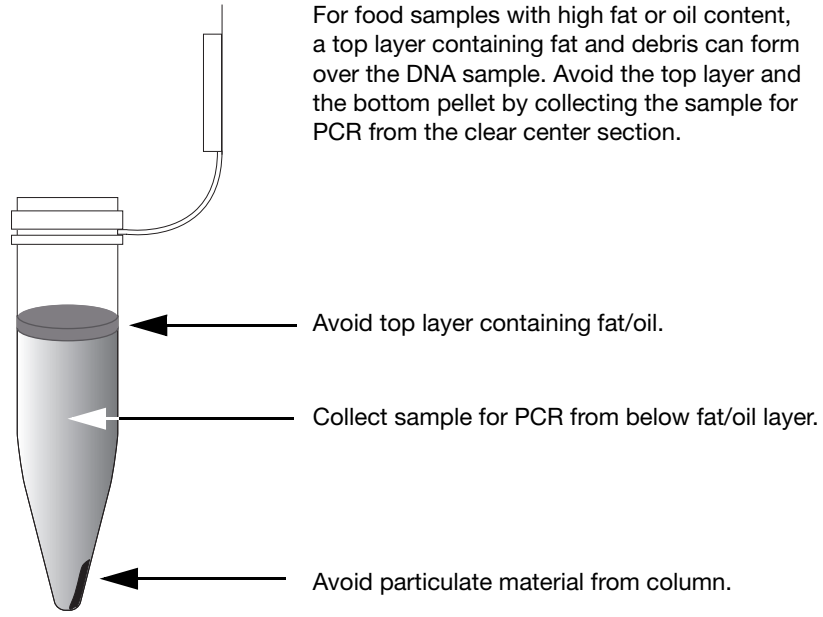
14. Microcentrifuge the tube at 12,000–16,000 × *g* for approximately 1 minute to bring down any particulate material derived from the spin column, which can interfere with amplification. The microbial DNA is in the aqueous phase.

15. Proceed with PCR, or store the tube below –18°C.

**IMPORTANT!** Follow these tips, when analyzing your DNA samples:

- For PCR, add 30 µL of supernatant to the lyophilized assay. For detailed instructions, refer to the *MicroSEQ® E. coli O157:H7 Detection Kit User Guide* (Pub. no. 4426511).
- Food samples with high fat or oil content can form a top layer containing fat and debris over the DNA sample. When collecting your sample for PCR, avoid the top layer and bottom pellet by collecting your sample from the clear center section (see the following figure).
- If PCR inhibition occurs, as indicated by no amplification of both target and IPC, then dilute sample with water. We recommend adding 5 µL of sample and 25 µL of water to the lyophilized assay to overcome inhibition. See "Troubleshooting" on page 17 for further discussion.

**To pipet eluate from foods with high fat or oil content:**



# Troubleshooting

**For food testing**

| Observation | Possible cause | Recommended action |
|---|---|---|
| Inhibition of PCR is indicated by non-detection of IPC reaction. | Removal of the sample supernatant before addition of Lysis Buffer was incomplete. | Add 5 µL of sample and 25 µL of water to the lyophilized assay to overcome inhibition. |
| | Filtrate from the spin column is in the PCR. | Centrifuge the sample to separate the filter particulates before adding sample to the PCR reaction. Pipet your sample from the clear center section, avoiding the particulate material at the bottom of the tube. |
| | The sample contained excess fat that was not removed during aspiration of the supernatant. | Apply PrepSEQ® Rapid Spin Extra Clean protocol when processing a matrix with high fat content. |
| | The matrix is associated with PCR inhibitory components. | Pre-wash the bacterial pellet before column clarification:<br>1. Transfer 750 µL of sample to a clean microcentrifuge tube.<br>2. Centrifuge at 12,000–16,000 × *g* for approximately 3 minutes.<br>3. Discard the supernatant.<br>4. Resuspend the pellet in 650 µL of sterile distilled water.<br>5. Load column and centrifuge. Continue with sample preparation step 3 on page 13. |
| The bacterial pellet separates from the tube, making the pellet difficult to avoid during aspiration. | The sample was left unattended before aspirating off the supernatant, causing dissipation of the bacterial pellet. | Re-centrifuge and remove the supernatant immediately following centrifugation. |

# Background Information

## Overview

Use the PrepSEQ® Rapid Spin Sample Preparation Kit to prepare food samples to test for *E. coli* O157:H7. The kit is designed for preparation of DNA from most food types. The kit procedure involves:

- Enrichment of food samples for *E. coli* O157:H7
- Sample preparation

For sample preparation from enriched food samples, we recommend a 750-µL sample volume.

For some foods with a high lipid content [such as infant formula, whole milk, smoked salmon (lox), and chicken wing samples], use the PrepSEQ® Rapid Spin Sample Preparation Kit – Extra Clean (Cat. no. 4413269). The PrepSEQ® Rapid Spin Extra Clean protocol is also recommended with some 375-g food samples depending on the sample's fat content.

## AOAC Performance Tested Methods℠ Certification

The MicroSEQ® *E. coli* O157:H7 Detection Kit earned the Performance Tested Methods℠ Certification from the AOAC Research Institute. The validation was conducted using USDA MLG 5.04 as the reference method for meat products, and ISO 16654 as the reference method for leafy green products and juices. The validated workflow includes:

- Two sample preparation kit options:
  - PrepSEQ® Nucleic Acid Extraction Kit
  - PrepSEQ® Rapid Spin Sample Preparation Kit
- MicroSEQ® *E. coli* O157:H7 Detection Kit
- Applied Biosystems® 7500 Fast Real-Time PCR Instrument
- RapidFinder™ Express Software

The method was certified for use with the following matrices:

- 25 g of ground beef and beef trim
- 375 g of ground beef and beef trim
- 25 g of spinach
- 25 g of apple juice
- 25 g of orange juice

# ISO 16140 Validation


**ABI 29/03 – 03/11**
[End of validity: refer to certificate at www.afnor-validation.com]

ALTERNATIVE ANALYTICAL METHODS FOR AGRIBUSINESS
Certified by AFNOR Certification

The MicroSEQ® *E. coli* O157:H7 Detection Kit was certified "NF Validation". The ISO 16140 standard was used for the validation of alternative methods. This kit was compared and found equivalent to the ISO 16654 reference method. The validated workflow includes:

- Two sample preparation kit options:
  - PrepSEQ® Nucleic Acid Extraction Kit
  - PrepSEQ® Rapid Spin Sample Preparation Kit
- MicroSEQ® *E. coli* O157:H7 Detection Kit
- Applied Biosystems® 7500 Fast Real-Time PCR Instrument
- RapidFinder™ Express Software

The method, using workflows A or B, was certified for use with the following matrices: raw beef meat and raw produce.

General recommendations:

- Comply with Good Laboratory Practices – GLP (Refer to EN ISO 7218 standard).
- In the context of NF Validation, sample sizes of more than 25 gram have not been tested.
- ISO 16654 and ISO 6887 are recommended for preparation of master suspension.

For more information about the expiration date of the "NF Validation" certification, please refer to the certificate, available on the website at: **www.afnor-validation.com**

# 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 Safety Data Sheet (SDS) for specific precautions and instructions:

- Read and understand the 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. Follow all applicable local, state/provincial, and/or national regulations. Wear appropriate protective equipment, which includes but is not limited to: protective eyewear, face shield, clothing/lab coat, and gloves. All work should be conducted in properly equipped facilities using the appropriate safety equipment (for example, physical containment devices). Individuals should be trained according to applicable regulatory and company/institution requirements before working with potentially infectious materials. Read and follow the applicable guidelines and/or regulatory requirements in the following:

In the U.S.:

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

In the EU:

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

## Obtaining SDSs

Safety Data Sheets (SDSs) are available from **www.lifetechnologies.com/support**.

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

## Obtaining Certificates of Analysis

The Certificate of Analysis provides detailed quality control and product qualification information for each product. Certificates of Analysis are available on our website. Go to **www.lifetechnologies.com/support** and search for the Certificate of Analysis by product lot number, which is printed on the box.

## Obtaining support

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

**www.lifetechnologies.com/support**

At the website, you can:

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

# Food safety support

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

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

**Phone number** (In North America): 1-800-500-6885

**Phone number** (Outside North America): Go to **www.lifetechnologies.com/contactus.html** and select the appropriate country from the drop-down menu.

# Limited product warranty

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