A Worldwide Healthcare Problem

One of the most common causes of foodborne illness is *Salmonella*, a genus of bacteria responsible for an infection called salmonellosis. Salmonellosis is a serious public health concern throughout the world. It is also a major concern for food growers, manufacturers, packers, shippers, wholesalers, and retailers. Release of contaminated foods can result in processing delays, product recalls, plant shutdowns, and legal action. It can cost food companies millions in revenue as well as their reputation, stock value, and future profitability.

The Testing Challenge

A key to preventing *Salmonella* contamination is timely and accurate food pathogen testing. *Salmonella* testing can be difficult due to the number of species and variety of foods in which the bacteria are found. To be effective, a *Salmonella* test must detect low levels of the bacteria in a wide range of food types quickly, easily, and with high specificity. The MicroSEQ® *Salmonella* spp. Detection Kit has been designed and optimized to provide such a solution.

Traditionally, *Salmonella* testing has been performed using culture-based or immunoassay methods, which are time-consuming and labor-intensive. With culture-based methods, for example, the time-to-results can take up to 5 days, and interpretation of the results is highly subjective. Immunoassay-based methods can take up to 2 days and may lack specificity due to antibody binding interferences.

By contrast, time-to-results with the MicroSEQ® *Salmonella* spp. Detection Kit is less than 2 hours after a one-step preenrichment, and less than 18 hours total. Results are highly specific and sensitive due to real-time PCR detection, so you can be confident in their accuracy.
Benefits of Real-Time PCR

The MicroSEQ® Salmonella spp. Detection Kit is based on real-time PCR. Real-time PCR is a proven method for pathogen detection and has been applied successfully to a wide range of foodborne pathogens, including Salmonella, Listeria monocytogenes, and E. coli O157:H7. The method tests for food pathogens by amplifying and detecting a DNA target sequence that is specific to the organism under investigation. In this way, real-time PCR can detect pathogens quickly, simply, and with outstanding sensitivity and specificity. And since the kit uses fast PCR chemistry, the PCR step takes only 40 minutes, compared with up to 2.5 hours using standard PCR.

A Complete Solution

The MicroSEQ® Salmonella spp. Detection Kit is part of a complete food-testing solution. The kit includes everything required to run 96 reactions. All components have been designed for rapid implementation in food-testing laboratories and performance-verified to make Salmonella detection as fast, easy, and reliable as possible. Everything is provided ready to use.

- Optimized for Sensitivity and Specificity: The assay is designed to provide maximum sensitivity on the 7500 Fast Real-Time PCR System.
- Ready to Use: The active enzyme, reagents, primers, probes, and internal positive control are lyophilized into preformatted assay beads. No mixing is required.
- Optimized Sample Preparation: A choice of sample preparation kits helps ensure high-quality results.
- Software-Guided: Application-specific RapidFinder™ Express Software guides the user through each step of the procedure—from run file setup to final results.
- AOAC-Certified: The full workflow including the MicroSEQ® Salmonella spp. Detection Kit, PrepSEQ™ Sample Preparation Kits, 7500 Fast instrument, and RapidFinder™ Express Software has earned the Performance Tested Methods™ certification from the AOAC Research Institute.

High Sensitivity and Specificity

Sensitivity, specificity, and robust performance are standard requirements for effective Salmonella testing. The bacteria can grow in virtually any raw food, including meat and dairy products, eggs, vegetables, and fruit. It has also been found in peanut butter, infant formula, salad dressings, seafood, dog food, spices, and chocolate. To further compound the problem, there are over 2,400 serotypes of Salmonella.

The MicroSEQ® Salmonella spp. Detection Kit detects 99% of all Salmonella serotypes. In combination with Applied Biosystems sample preparation kits, the kit can detect 1–3 colony forming units (cfu) per 25 grams of sample, with a limit of detection of $10^{3}$ cfu/mL. This level of performance has been demonstrated in a wide range of foods, including complex food types such as chocolate and infant formula.

Ease of Use and Reliability

The MicroSEQ® Salmonella spp. assay can be performed in 3 simple steps by nontechnical personnel with minimal training or prior experience. For maximum ease of use, stability and reliability, and consistency of results, the reagents used in the Salmonella assay are preformatted and lyophilized into assay beads. The beads hold the active enzyme, the target-specific primer and probe set, internal positive control (IPC), and other reagents for PCR. The IPC is provided to help eliminate false negatives by detecting the presence of materials that can inhibit target amplification.

There is no electrophoresis or post-PCR processing. All the operator has to do...
is to prepare the assay beads, add the samples and controls, and run the PCR test. Sample handling is minimal, and every step is guided by the RapidFinder™ Express Software with on-screen instructions.

For added reliability, the MicroSEQ® Salmonella spp. Detection Kit uses specially designed reaction tubes that remain closed throughout the assay process. Once the sample is added, the tubes are closed and remain that way until detection is complete, greatly reducing the chances of contamination.

**Optimized Sample Preparation**

The MicroSEQ® Salmonella spp. Detection Kit is optimized for use with the PrepSEQ™ Sample Preparation Kits. The PrepSEQ™ kits remove PCR inhibitors, enhance assay performance, and enable the user to enrich and prepare high-quality samples from a broad range of food types for detection of 1–3 cfu per 25 grams of sample.

- For automated high-throughput applications, the PrepSEQ™ Nucleic Acid Extraction Kit prepares high-quality microbial DNA and RNA from broth cultures when used with the Applied Biosystems MagMAX™ Express-96 Sample Preparation System.
- For lower sample numbers, the PrepSEQ™ Rapid Spin Sample Preparation Kit provides a simple, cost-effective way to prepare DNA from broth cultures.

**Fast, Actionable Answers**

When the assay is complete, RapidFinder™ Express Software presents an easy-to-read screen that allows the user to view the results in each reaction location. Results are clearly displayed and can be labeled with flags, notifications, and prompts that enable the operator to quickly interpret the data and take appropriate action.

**Resources for Food Safety**

As the world leader in real-time PCR, Applied Biosystems is committed to providing the food industry with improved tools for pathogen detection. The MicroSEQ® Salmonella spp. Detection Kit is part of a growing family of fast and convenient food pathogen detection tools that utilize lyophilized reagents, application-specific software, optimized sample preparation, and fast real-time PCR instrumentation. Other solutions in our expanding portfolio include easy-to-use TaqMan® Pathogen Detection Kits for rapid detection of a broad menu of food pathogens. We also provide responsive, knowledgeable applications consulting, support, training, and technical service.

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**Figure 2. The MicroSEQ® Salmonella spp. Detection Kit produces fast, accurate results in three simple steps.**

1. **Enrich Samples**
2. **Prepare Samples**
3. **Run Test**

Using either the PrepSEQ™ Nucleic Acid Extraction Kit or the PrepSEQ Rapid Spin Sample Preparation Kit.

According to RapidFinder™ Express Software instructions.

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**Figure 3. Software-Guided Procedure.** After sample enrichment and preparation, RapidFinder™ Express Software guides the user through the entire workflow with on-screen instructions. Amplification, detection, data collection, and analysis are fully automated.
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For more information about the MicroSEQ® Salmonella spp. Detection Kit and our other solutions for food pathogen testing, please contact your local Applied Biosystems sales representative or visit us at www.appliedbiosystems.com/foodsafety.

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MicroSEQ® *Listeria monocytogenes* Detection Kit

Fast, Accurate Food Pathogen Testing

- Easy-to-use, streamlined lyophilized format
- Fast time-to-results
- High sensitivity and specificity for confidence in results
- Optimal performance with wide range of food sample types
- Internal positive control helps eliminate false negatives

**A Dangerous Foodborne Pathogen**

Tasteless, odorless, and impossible to see with the naked eye, *Listeria monocytogenes* is responsible for listeriosis, a disease with a high fatality rate in susceptible populations, including newborns, immunocompromised individuals, and the elderly, causing the highest mortality rate among foodborne illnesses. Listeriosis is particularly dangerous for pregnant women, potentially causing miscarriage or stillbirth.

**A Concern for the Food Industry**

In addition to its impact on health, an outbreak of listeriosis can be devastating to food processors. If the outbreak is traced to a facility, the operator may be forced to recall and destroy contaminated products, shut down operations, and face lost revenue, damaged reputation, fines, and litigation. Containment is complicated by the fact that *Listeria* can grow at temperatures as low as 3°C, allowing it to multiply in refrigerated foods during storage, shipping, and retail display.

**The Testing Challenge**

*Listeria monocytogenes* is one of six species of the genus *Listeria* and the only identified species to cause listeriosis. *Listeria monocytogenes* bacteria grow slowly and are difficult to distinguish from other species of *Listeria* on agar culture media. This has prompted many food-testing laboratories to search for a test method that provides rapid, accurate results—with high sensitivity and specificity—that is easy to use and implement. The MicroSEQ® *Listeria monocytogenes* Detection Kit meets these criteria.

**Benefits of Real-Time PCR**

Real-time PCR is a proven method for pathogen detection and has been applied successfully to a wide range of foodborne pathogens, including *Salmonella*, *Listeria monocytogenes*, and *E. coli* O157:H7. Real-time PCR tests for food pathogens by amplifying and detecting a DNA target sequence that is specific to the organism under investigation. In this way, real-time
PCR can detect pathogens quickly, simply, and with outstanding sensitivity and specificity. And since the kit uses fast PCR chemistry, the PCR step takes only 40 minutes, compared with up to 2.5 hours using standard PCR.

**Improved Time-to-Results**

Real-time PCR offers faster time-to-results than other methods. *Listeria* testing using traditional culture-based methods can take up to 5 days. Immunoassay-based methods take up to 2 days. Time-to-results with the MicroSEQ® *Listeria monocytogenes* Detection Kit is less than 3 hours after preenrichment, and less than 27 hours total.

**High Specificity and Sensitivity**

Because the MicroSEQ® *Listeria monocytogenes* Detection Kit detects genetic material unique to the organism, it provides both high specificity and sensitivity. With culture-based methods, interpretation of the results is highly subjective, which may lead to missed or ambiguous identification of *Listeria monocytogenes*. Immunoassay methods, while less subjective than culture methods, are based on antibody-binding mechanisms that may be prone to interference and lead to false positives and/or false negatives.

The MicroSEQ® *Listeria monocytogenes* Detection Kit specifically detects the following *Listeria* serotypes: 1/2A, 1/2B, 1/2C, 3A, 3B, 3C, 4A, 4AB, 4B, 4C, 4D, and 7. The kit does not detect other pathogens.

In combination with the PrepSEQ™ sample preparation protocols, the kit can detect 1–3 colony forming units (cfu) per 25 grams of sample, with a limit of detection of $10^3$ cfu/mL.

**Lyophilized for Efficiency and Ease of Use**

For maximum ease of use, reliability, and consistency of results, the reagents used in the MicroSEQ® *Listeria monocytogenes* Detection Kit are lyophilized into preformatted assay beads. The beads hold the active enzyme, the target-specific primer and probe set, internal positive control (IPC), and other reagents for PCR. The IPC is provided to help eliminate false negatives by detecting the presence of materials that can inhibit target amplification.

**Closed-Tube Integrity**

The MicroSEQ® *Listeria monocytogenes* Detection Kit uses specially designed reaction tubes that remain closed throughout the assay process. Once the sample is added, the tubes are closed and remain that way until detection is complete, greatly reducing the chances of contamination.

No electrophoresis or post-PCR processing is required. All the operator has to do is to prepare the assay beads, add the samples and controls, and run the test. Sample handling is minimal, and every step is guided by the RapidFinder™ Express Software with on-screen instructions.

**Optimized Sample Preparation**

The MicroSEQ® *Listeria monocytogenes* Detection Kit is optimized for use with the PrepSEQ™ Sample Preparation Kits. The PrepSEQ™ kits remove PCR inhibitors, enhance assay performance, and enable the user to enrich and prepare high-quality samples from a broad range of food sample types for detection of 1–3 cfu per 25 grams of sample.

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A Complete Solution

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- **Optimized Sample Preparation:** A choice of sample preparation kits helps ensure high-quality assay results.
- **Software-Guided:** Application-specific RapidFinder™ Express Software guides the user through each step of the procedure—from run file setup to final results.
- **AOAC-Certified:** The full workflow including the MicroSEQ® Listeria monocytogenes Detection Kit, PrepSEQ™ Sample Preparation Kits, 7500 Fast Instrument, and RapidFinder™ Express Software has earned the Performance Tested Methods℠ certification from the AOAC Research Institute.

Resources for Food Safety

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For more information about the MicroSEQ® Listeria monocytogenes Detection Kit and our other solutions for food pathogen testing, please contact your local Applied Biosystems sales representative or visit us at www.appliedbiosystems.com.

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**Figure 3.** The MicroSEQ® Listeria monocytogenes Detection Kit streamlines the assay workflow with optimized sample preparation and assay procedures. Nontechnical personnel with minimal training or prior experience can perform the assay in 3 simple steps.

**Figure 4.** Software-guided procedure. After sample enrichment and preparation, RapidFinder™ Express Software guides the user through the entire workflow with on-screen instructions. Amplification, detection, data collection, and analysis are fully automated.
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### Sample Preparation

- PrepSEQ™ Nucleic Acid Extraction Kit 4400799
- PrepSEQ™ Rapid Spin Sample Preparation Kit with Proteinase K 4426714
- PrepSEQ™ Rapid Spin Sample Preparation Kit – Extra Clean with Proteinase K 4426715

### Instrumentation and Software

- Food Pathogen Detection System Package: Applied Biosystems 7500 Fast Real-Time PCR System with PC Tower and RapidFinder™ Express Software 4445785
- Food Pathogen Detection System Package: Applied Biosystems 7500 Fast Real-Time PCR System with Notebook Computer and RapidFinder™ Express Software 4445787

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