Listeria spp., Fostering a shift in food safety culture





Welcome!



Shawn Gartside Regional Sales Manager shawn.gartside@sample6.com



Trevor Morones Lead Food Safety Consultant tmorones@controlpoint.consulting

Topics

- Characteristics
- History
- Presence
- Environmental monitoring
- Food safety culture
- Sampling procedures/collection
- Available technologies.
- Testing
 - In-house vs. 3rd party
- Methods
 - Costs, features, and benefits



What area of the food and beverage business do you represent?

Retail?

Animal/Pet Supplier Manufacturer?

Domestic Manufacturer?

Distribution?

Produce - Grower/Packer/Processor?



Listeria

- 1924 isolated <u>Gram-</u> <u>positive</u>rods
- 1940 named genus Listeria for <u>catalase-</u> <u>positive, Gram-</u> <u>positive rods</u>
- 1949 Epidemic of listeriosis in newborns in Germany



Lm – Listeria *monocytogenes*

- Gram-positive
- Catalase-positive
- Non-sporeforming
- Slim rod
- Colonies are smooth, convex, circular, bluish translucent appearance on nonselective agars; atypical rough colonies form filaments



Outbreak



rachelcooks.com

- First Outbreak Canada 1981, Coleslaw
- 1600 illnesses and 260 deaths due to listeriosis occur annually in the United States
- Serious threat to high-risk populations

Presence of Listeria

- Entry
 - Soil (shoes, vehicles, clothing)
 - Contaminated raw plant and animal tissue
 - Human carriers
- Popular locations
 - Floor drains
 - Condensed water
 - Floors
 - Processing equipment
 - Surfaces





"As we look ahead into the next century, leaders will be those who empower others."

William H. Date I



What is the biggest challenge you face with food safety culture?

- Management support?
- Upper management support?
- Tools to deliver the message?
- How to Start?

Food Safety Culture

- Knowledge is power
- A strong culture becomes contagious
 - Trends are contagious
- Every team member has a responsibility to ensure safe unadulterated food enters commerce



From the start

- Culture should be instilled from day one
 - Onboarding
 - Uniform
 - Physical environment
 - Break areas
 - Locker rooms
 - Restrooms
 - Management style

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News: FSIS Pilot - Notice 04-16 1/20/16



1/20/16

PILOT PROJECT: CONTROL OF LISTERIA MONOCYTOGENES (LM) IN RETAIL DELICATESSENS

I. PURPOSE

On January 25, 2016, FSIS will launch a year-long pilot project to assess whether retailers are using the recommendations in the *FSIS Best Practices Guidance for Controlling Listeria monocytogenes (Lm) in <u>Retail Delicatessens</u> (FSIS Retail <i>Lm* Guideline). This notice provides instructions to the Office of Investigation, Enforcement and Audit (OIEA), Compliance and Investigations Division (CID) Investigators, on how to complete the Ready-To-Eat (RTE) Retail Deli Tool, a questionnaire in the Public Health Information System (PHIS) that will help Investigators compare the practices observed in retail delis to the FSIS Retail *Lm* Guideline in the following areas: product handling, cleaning and sanitizing, facility and equipment controls, and employee practices. This notice also provides instructions to Investigators on how to conduct outreach at retail firms that slice or prepare deli products during their in-commerce surveillance activities.

pilot project to assess
whether retailers are using
the recommendations in the
FSIS Best Practices
Guidance for Controlling
Listeria *monocytogenes* (Lm)
in Retail Delicatessens (FSIS
Retail Lm Guideline)

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Fostering a shift in food safety culture

- Abbott, Cheese British Columbia (2002)
- Whittier Farms, Pasteurized Milk – Massachusetts (2007)
- Sangar, Fresh Cut Produce
 Texas (2010)
- Jensen Farms, Cantaloupe– Nationwide (2011)
- Marte, ricotta salata cheese– Multistate (2012)



Environmental Monitoring

- interested in finding any type of Listeria (generic Listeria species).
 - Non-food contact surfaces and food contacts surfaces
- Best practice to hold lots until results are received but not always feasible



Key words and concepts

Listeria sp. vs Listeria monocytogenes-

- <u>Listeria species-</u>contain several types of Listeria, not regulated. Most companies use as indicator for environmental testing.
- <u>Listeria monocytogenes</u>- Specific Listeria species that is pathogenic with a zero tolerance regulation in food.

Environmental testing vs. Product testing- Proactive (HACCP) vs. Reactive

- Presumptive Positive Any positive result by a rapid method that is not USDA or FDA culture
- Confirmation- USDA or FDA culture, or secondary verification by another technology. Listeria sp. tests are sometimes confirmed for Listeria mono.
- Enrichment Media- Liquid or solid nutrients for the growth of bacteria-sometime specific for the type of bacteria
 - UVM, BLEB, Frasier broth, MOX
- False positive and False negative- measurement of the accuracy of a test. Determined by confirmation of original sample

Listeria testing methods-Characteristics

	PCR	Immunoassay	Sample6
Technology base	DNA or RNA	Cell Surface- Antibodies and Antigens	Phage based with Bioluminescent detection
Time to result	 18-48 hour growth 1-4 hours detection 20-52 hours TTR 	 30-48 hours growth 10 minutes-2 hour detection 30-50 hours TTR 	 6 hour resuscitation 5 minute detection 7 hour TTR
Detection limit	1 CFU pre-enrichment 10,000 CFU/mL post	1 CFU pre-enrichment 100,000+ CFU/mL post	1 CFU pre-incubation
Software	Included	Some times Included	Included

Are you currently testing for Listeria?

- In-House?
- 3rd Party Lab?
- Both
- No?



3rd party vs. in house testing

	3 rd party lab	In house lab
Location	Off site away from plant	At or near plant- consider method carefully
Time to result	2-5 days	7-48 hours
Cost	\$20-\$55 / test plus overnight shipping or courier	\$8-\$30 / test
Regulatory	Certified and Use AOAC methods Some AOAC methods not USDA approved	Document training and GLP Some methods not USDA approved

Listeria testing methods- Capital equipment

	PCR	Immunoassay	Sample6
Equipment Cost	\$15,000-\$55,000 (includes thermocycler, computer workstation, monitor)	\$35,000 (liquid handler, computer, hand-held scanner)	\$10,000 (includes luminometer, centrifuge, pipette, iPad, other supplies)
Additional Equipment Needed	Heat Blocks with insert: \$1,800-2,000 Incubator	Heat Block: \$800 Incubator	Standard Incubator
Maintenance Contract	Included in Contract	\$5,000-\$7,000 / year	Not Needed



Comparing costs-Test and consumables

	PCR	Immunoassay	Sample6
Kit Cost	\$650-\$950 (96 tests)	\$300 -\$600 (60 tests)	\$1,600 (100 tests)
Enrichment Media Costs	\$60 - \$80 (96 tests)	\$80 - \$100 (60 tests)	Included
Lab Supply Costs	\$100 - \$200 (96 tests)	\$100 - \$200 (60 tests)	Included
Total Cost per Test	\$9 - \$11	\$8 - \$15 per test	\$16 per test

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Comparing costs-Labor considerations

	PCR	Immunoassay	Sample6
Operator Skill	Bachelor or Masters + Experience	Bachelor or Masters + Experience	No Technical Degree Required
Sample Prep Time	2 - 3 minutes / sample	2 - 3 minutes / sample	2 - 3 minutes / sample
Hands-on Analyst Time	35 - 40 minutes	60 - 90 minutes for one run	15 minutes
Incubation Time	48 hours	28 - 32 hours total	6 hours
Assay Time	3.5 hours	90 minutes	20 seconds/sample



Transform Pathogen Food Testing

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Integrated, End-to-end Solution for Environmental Food Testing



AOAC Certification & USDA Authorized

AOAC

- The Association of Official and Analytical Chemists (AOAC) is an independent nonprofit organization that was formerly tied to the FDA, whose responsibilities are to:
 - Establish standards
 - Conduct conformity assessment

The food industry de facto requires that any new assay be AOAC-certified before food processors evaluate and commit to deployment

Sample6 Certification

AOAC-RI PTM (Performance Tested Method)

MICROBIOLOGICAL METHODS

Sample6 DETECT/L: An In-plant, In-shift, Enrichment-free Listeria Environmental Assay

Performance Tested MethodSM 041401

THOMAS HAMMACK

The Sample6 DETECT/LTM Kit is a rapid screening assay that utilizes proprietary Sample6 BioilluminationTM technology for the detection of Listeria species on environmental surfaces. The assay was evaluated according to AOAC guidelines for inclusivity, exclusivity, robustness, instrument variation, and product consistency and stability, as well as in a method comparison study using one environmental surface. The validation was conducted using an unpaired study design for the detection of Listeria species on stainless steel surfaces. Statistical analyses were conducted according to the probability of detection statistical model for the robustness, stability, instrument variation and method comparison studies. The Sample6 DETECT/L assay correctly identified all 50 inclusivity isolates and correctly excluded all 30 nontarget strains evaluated in the inclusivity and exclusivity studies. The robustness study demonstrated that the assay was not affected by variations in incubation temperature, incubation time, or the volume of the detection reagent substrate. Regarding instrument variation, no differences were observed between replicates analyzed using three different instruments. In the stability study consistent results were obtained from three different product lots spanning the shelf life of the assay. The results of the statistical analysis of the method comparison indicated there was no significant difference between the Sample6 DETECT/L method and the reference method with the exception of sensitivity where the Sample6 method was superior to the reference.

Participants

METHOD AUTHORS MICHAEL CAPPILLINO, ROBERT P. SHIVERS, DANIEL R. BROWNELL, BECK JACOBSON, JOHN KING, PAULINA KOCJAN, MICHAEL KOERIS, ED TEKEIAN, ANDREW TEMPESTA, and JAYSON BOWERS Sample6, 840 Memorial Dr, Cambridge, MA 02139

ERIN CROWLEY, PATRICK BIRD, JOE BENZINGER, and KIEL FISHER Q Laboratories, Inc., Cincinnati, OH 45214

SUBMITTING COMPANY Sample6, 840 Memorial Dr. Cambridge, MA 02139

INDEPENDENT LABORATORY Q Laboratories, Inc., Cincinnati, OH 45214

Submitted for publication October 15, 2014. The method was independently tested, evaluated, and certified by the AOAC Research Institute as a *Performance Tested Method^{3M}*. See http://www.aoac.org/testkits/steps.html for information on certification. ^{*} Corresponding author's e-mail: jayson.bowers@sample6.com DOI: 10.5740/jaoacint.14-213

REVIEWERS

U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition, 5100 Paint Branch Pkwy, College Park, MD 20740

MICHAEL BRODSKY

Brodsky Consultants, 73 Donnamora Crescent, Thornhill, Ontario L3T 4K6, Canada

FUTIOT RUSER Michigan State University, 2108 Anthony Hall East, Lansing, MT 48824

Scope of Method

(a) Target organisms.-Listeria species (L. monocytogenes, L. innocua, L. ivanovii, L. seeligeri, L. welshimeri, and L. marthii).

(b) Matrix.-Stainless steel environmental surfaces.

(c) Performance claims .- Sensitivity equivalent to the U.S. Department of Agriculture/Food Safety and Inspection Service-Microbiology Laboratory Guidebook (MLG) 8.09 (1) reference culture method based on probability of detection (POD) statistical analysis. Correct identification of all 50 target strains and correct exclusion of all 30 nontarget strains. No statistically significant differences were observed in the robustness, instrument variation, and stability studies.

(d) Probability of detection .- POD is the proportion of positive analytical outcomes for a qualitative method for a given matrix at a given analyte level or concentration. POD is concentration-dependent. The difference in POD (dPOD) is the difference between any two POD values. If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level.

Introduction

Listeria species are Gram-positive, non-spore forming aerobic organisms that are ubiquitous in the environment. Until the 1960s, infections caused by Listeria were thought to be associated almost exclusively with animals and less frequently or rarely with humans. In subsequent years, Listeria species, specifically L. monocytogenes, began to be isolated from a wide range of hosts, including humans (2). During the 1980s, L. monocytogenes emerged as an important foodborne human pathogen because of its persistence in many food processing environments and ability to grow at refrigeration temperatures. This foodbome pathogen has since been isolated from a wide range of foods including milk, cheeses, delicatessen meats, hot dogs, smoked fish, and most recently fresh produce (3). Pregnant women, the elderly, and immunocompromised individuals are

Method Published in the Journal of AOAC International

How do you currently manage your testing results?

Excel spreadsheet?

Lab Notebook?

Other software?

I don't know?



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EASIEST

EASY

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0 hours

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Engineered phage interacts specifically with target bacterial cells Bacteria over-produce the **reporter enzyme**

10K ENZYMES

Cells **lyse** and the reporter is detected

100M PHOTONS

6 hours

1 CELL

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DETECT[™] & CONTROL[™]



FSMA: Food Safety Program & Sample6 What does it mean and how can we help



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