Understanding Microbiological Sampling and Testing

FSIS 2016 EIAO Education Program

Danah Vetter, DVM
Veterinary Medical Officer
Office of Public Health Science
Science Staff
Food Safety and Inspection Service:

Today’s Presentation

• FSIS and industry testing activities
• Sampling methods and design
• Testing methods
  – Fitness for purpose
  – Validation
  – Issues specific to pathogen testing
  – Quantitative testing
• Laboratory accreditation and communications
Food Safety and Inspection Service:

FSIS and Industry Testing Programs
Food Safety and Inspection Service:

**Microbiological Testing by FSIS Laboratories**

- Three Field Service Labs administer regulatory testing programs
  - Athens, Georgia
    - Executive Associate
    - EFSL-routine/other testing
    - LQAD-quality assurance
    - FERN- biosecurity
  - St. Louis, Missouri
    - MWFSL-routine testing
  - Alameda, California
    - WFSL-routine testing
- Routine monitoring, follow-up, baseline study programs and investigative sampling

ISO 17025 Accredited
### FSIS Microbiological Sampling Programs

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>No. Collected</th>
</tr>
</thead>
<tbody>
<tr>
<td>Domestic</td>
<td>75,346</td>
</tr>
<tr>
<td>Import</td>
<td>7,482</td>
</tr>
<tr>
<td>In Commerce</td>
<td>485</td>
</tr>
</tbody>
</table>

**Fiscal year 2015 data**
Food Safety and Inspection Service:

**FSIS Micro Sampling Program Objectives**

- Assess effectiveness of industry process controls
- Provide critical feedback to industry
- Monitor compliance with performance standards, zero tolerance policies
- Allow FSIS to monitor industry-wide trends
- Serve as a strong incentive to reduce the occurrence of pathogens in products
- Capture pathogen subtyping information (i.e. serotype, PFGE pattern, antimicrobial resistance, whole genome sequencing)
Food Safety and Inspection Service:

**FSIS Sampling Programs**

- Sampling plans measure compliance with performance standards:
  - *Salmonella* and *Campylobacter j/c/l* verification programs (400 mL rinsate, 325 grams of comminuted poultry, 2 x 50 sq cm sponge)
- Zero tolerance policies for food pathogens
  - *E. coli O157:H7* (325 grams raw product)
  - Non-O157 Shiga toxin producing *E. coli* (325g N60 trim sample)
  - *Lm* (25 grams RTE product, presence in food contact surface swab, 25 grams pasteurized egg product)
  - *Salmonella* (325 grams RTE product or 100 grams pasteurized egg product)
Food Safety and Inspection Service:  
**Microbiological Testing by FSIS-Regulated Establishments (Industry testing)**

- Fulfill regulatory requirement (9 CFR 310.25, 381.94, 430.4, 590.580)
- Support decisions made in hazard analysis (9 CFR 417.2 (a))
- On-going verification of HACCP plan (9 CFR 417.4 (a)(2))
- Evaluate effectiveness of sanitary SOPs (9 CFR 416.14)
- Fulfill purchase agreements
- Respond to process deviations
The establishment is ultimately responsible for the testing they request from private laboratories.

Has the establishment properly conveyed testing needs?
- *e.g.*, test portion equivalent to FSIS as opposed to the default 25g in protocols.

Is the laboratory aware of FSIS expectations?
- Directives, Notices and guidance

Establishment should provide documented detailed methodology and validation information for FSIS review.
Food Safety and Inspection Service: 
**FSIS Verification of Establishment Sampling and Testing Programs**

- Effectiveness verified by FSIS
  - Review/Observations of EIAOs during FSA
  - Establishment provides supporting documentation
  - Technical and policy support provided through askFSIS
  - Establishment, not lab, is responsible for implementing effective program
Focus of FSIS’ evaluation
  – Is the method fit for the intended purpose?
  – Does the method support the hazard analysis decisions?
  – Is the method comparable to the appropriate FSIS method (or is there justification for an alternative)?
  – Is a comparable or appropriate test portion used?
  – Is the method validated and used under validated conditions?
  – Does the laboratory assure the quality of the results?
Assessing Sampling Plans
Why are Pathogens Hard to Detect?

- They are typically not evenly distributed
- They are often injured when found in the product
- They are able to cause disease at low levels
- Detection may be inhibited by material in the food product


Food Safety and Inspection Service:

**Sampling Methods and Tools**

- Destructive sampling (e.g., RTE, ground products, egg products)
- Non-Destructive sampling
  - Pro: when destructive sampling not an option
  - Examples:
    - Chicken carcass and chicken parts rinsate, carcass sponge samples
    - Food contact surface/Environmental sponge sample
Food Safety and Inspection Service:

**Sampling**

- All sampling plans have significant limitations
  - Therefore, we evaluate relative rigor of the program
- Best sampling plans provide the opportunity but no guarantee of detection
  - *i.e.*, scattered contamination is difficult to detect
- Frequent sampling and sampling multiple sites/time points provides a better opportunity for detection
  - Examples:
    - Multiple samples per day vs. once per month
    - “n60” per lot vs. one “grab” sample per lot
- Does the type of sampling meet the intended need?
  - Destructive vs. non-destructive sampling
Sampling Plans

- Statistical sampling plans assume
  - Uniform manufacturing conditions
  - Equal probability of contamination throughout the lot ("homogeneous distribution")
  - Independent, random sampling (equal probability of sampling throughout the lot)
Food Safety and Inspection Service:

*E. coli O157:H7* Contamination in a “n60” Sampled Lot

(illustration)
Food Safety and Inspection Service:

*E. coli O157:H7* Contamination in Ground Beef (illustration)

40% of product contaminated by hour 3 of production

Combo bins

40% of product contaminated by hour 3 of production

“slug”
What is “n60”? 

- “n60” = number of samples (n) = 60
  - Multiple representative samples provides best option for detecting scattered contamination
  - Provides 95% confidence that no more than 5% of food pieces the size of each “n” in the entire lot are contaminated

- Keys to success
  - Must ensure that sampling is as representative as possible across the lot
  - Large composite “n60” samples typical need a larger test portion
Common Sampling Problems

- Small sample or sampling method may not be ideal for detection
  - Examples: small swab device, small carcass or environmental area sampled
- Sanitizer or residual antimicrobial chemicals might interfere with the test
  - Insufficient drip time prior to carcass sample collection
  - Excessive liquid carryover for parts sample collection
- Temperature abuse for the sample prior to testing
  - Holding under refrigeration for long periods allows competing bacteria to grow
  - Freezing can kill some pathogens (e.g., *Campylobacter*)
Assessing Testing Methods
Food Safety and Inspection Service:

**Key Players for Ensuring Robust Testing Methods**

- The establishment that needs the testing
- The laboratory they hire
- The manufacturer of the screening test they use
- The organization validating the screening test
Steps in Detection Methods

- Sample collection
- Sample preparation
- Enrichment for the pathogen
- Screening of the Pathogen
- Confirmation of the Pathogen
Considerations for Testing Methods

- Is the method fit for the intended purpose of the analysis?
- Has the method been optimized and experimentally validated for sensitive detection of pathogens?
- Is the laboratory complying to the validated method protocol?
Assessing Fitness for Purpose

- Is the test portion appropriate to meet the need?
- Is the method enrichment-based with the intent to detect the lowest possible numbers of stressed pathogen cells?
- Are confirmation procedures appropriate for determining true negative samples?
The “Test Portion”

- Laboratory sample preparation => “test portion”
  - a.k.a., “analytical unit” or “analytical portion”
  - Definition- the part of the “sample” that is actually tested by the laboratory

- The test portion determines the theoretical (i.e., best possible) sensitivity of the test
  - i.e., 1 cell/test portion
  - 25-gram- detecting 0.04 cells/gram is possible
  - 325-gram- detecting 0.003 cells/gram is possible
Food Safety and Inspection Service:

**Enrichment**

- Test portion is incubated 8-48 hours in a culture broth
  - Why?
    - Contamination levels are too low for detection without enrichment
    - Must grow to high levels so very small volumes have enough for later detection steps.
  - Different pathogens require a different broths
  - One vs two-stage enrichment
    - resuscitation vs selective growth
Considerations for Proper Enrichment

• Resuscitation (lag phase) can require 2-3 hours before log-phase growth begins
  – Some samples support slower growth
• Has enrichment broth been tempered to warm temperature prior to incubation?
  – Particularly critical for large test portions or shorter incubation periods
**Pathogen Growth During Enrichment**

- **Log pathogen level (e.g., cfu, MPN/gram)**
- **Incubation time, hrs**
  - lag
  - logarithmic
  - stationary
  - death

**Possible Loss of Sensitivity Prior to confirmatory retesting**
• Different screening tests require different levels of enriched pathogen

• Shorter incubation periods (<15 hours) may warrant additional scrutiny of laboratory compliance to the validated protocol

• Has enrichment/screening combination been validated for a larger test portion?
  – Particular concern for large test portions incubated for shorter periods
    – e.g., 375-gram test portion incubated for 8 hours

• Proposed incubations < 8 hours may warrant OPHS review
Role of Enrichment
Food Safety and Inspection Service:

Validation of Methods
Food Safety and Inspection Service:

**Value of Validation**

- Determines performance characteristics of the method in comparison to a gold standard method (*i.e.*, usually FSIS or FDA method)
- Independent evaluation provides credibility
- Rigor varies (multilab vs. single lab, # tests, etc)
- Still must consider fitness for purpose and how the method is applied
  - *e.g.*, some AOAC-validated methods are not consistent with FSIS goals or Compliance Guidelines
Recognized independent method validation organizations:
- Government: FSIS (MLG) and FDA (BAM)
- AOAC International (U.S.A.)
  - AOAC Official Method (OM) validations
  - AOAC-RI “Performance Tested Method” validations
    - AFNOR (France)- e.g., bioMerieux-Vitek tests
    - Others (ISO, MicroVal, NordVal, etc.)

However, past validations conducted by these organizations may not be relevant to larger test portions or other testing scenarios.
Food Safety and Inspection Service:

Foodborne Pathogen Test Kits Validated by Independent Organizations

- FSIS maintains a list, updated quarterly, of methods that have been validated by independent organizations
- None of the test kits listed are implicitly approved by USDA FSIS
  - A validated test kit must also be fit for purpose and appropriate for the specific application in a food safety program
Food Safety and Inspection Service:

Process for Validating Qualitative Pathogen Methods

- Series of laboratory experiments using inoculated samples under controlled conditions
- Inoculate portions with pathogen strain at very low level where only 20-80% of samples are positive (i.e., fractional recovery)
- Statistically compare percent of positive samples in alternative method to reference method (FSIS MLG)
Considerations for Validation Data

• Was method compared to an appropriate reference method (e.g., FSIS MLG)?

• If not performed by AOAC, AFNOR, etc., is supplemental validation data available?
  – May require additional scrutiny
Testing Method Specifications

- **Sensitivity**: probability that truly positive samples are detected as positive by analytical test
  - 100 – false negative rate
- **Specificity**: probability that truly negative samples detected as negative by analytical test
  - 100 – false positive rate
- **Level of detection (LOD)**: lowest level of contamination reliably detected by analytical test
  - LOD expressed as ratio of organisms to quantity tested material (e.g., CFU per gram, MPN per mL, CFU per square-ft) but definitions vary (e.g., LOD50, POD)
Factors Impacting Detection and Method Specifications

• Detection as measured by sensitivity, specificity, and LOD can vary based on:
  – Specific strains of pathogen
  – Intrinsic factors for the sample matrix
    • Levels of competing bacteria
    • Fat, salt, pH and additives
  – Experimental design for the validation study (e.g., cell stress, etc.)
Confirmatory Testing

- Non-culture confirmation (e.g., PCR)
- Culture confirmation (e.g., FSIS confirmation)
  - Plating the enrichment on selective and differential agar media
  - Immunomagnetic separation (IMS) necessary prior to plating for *E. coli* O157:H7 and Non-O157 STECs
  - Suspect colonies = “presumptive positive”
- Purification and confirmatory identification tests including:
  - Biochemical (e.g., identifies “*E. coli*”)
  - Serological (e.g., identifies “O157” and “H7”)
  - Genetic (e.g., identifies “*stx*” = Shiga toxin genes)
Concerns for Confirmation

• Do not re-sample the lot or sample reserve!

• Non-culture confirmation
  – Same considerations as the screening test
  – Used under validated conditions
  – Transport and storage of enrichment

• Culture confirmation- carefully assess!
Complying with the Validated Protocol

• Do AOAC/AFNOR citations match the protocol in use?
  – Modifications are common, and some contribute to greater potential for false negative result

• Compare the lab procedure to the validated protocol

• If culture confirmation is used, verify that it follows validated method as well
Food Safety and Inspection Service:

**Methods Not Validated by Recognized Organizations**

- “Supplemental” or “extension” validations
- *E. coli* O157:H7 and Non-O157 STEC testing for 325-375g test portions.
  - Modifications required for AOAC validated procedures based on 25g
  - Instructions may not be clear for the lab
- Non-O157 STEC “No Objection Letter” process
Food Safety and Inspection Service:

FSIS Analytes
Food Safety and Inspection Service:

**STEC Testing**

- Includes:
  - *E. coli* O157:H7
  - Six Non-O157 Shigatoxigenic *E. coli*
Food Safety and Inspection Service:

**E. coli O157:H7 Analysis (MLG Ch. 5A)**

<table>
<thead>
<tr>
<th>Day 1</th>
<th>Sample Prep and Primary Enrichment 42°C±1 for 15-22 hours</th>
<th>Enrichment</th>
</tr>
</thead>
</table>
| Day 2 | Perform PCR  
All samples that do not test PCR negative are carried forward for further analysis | confirm (-), potential (+)  
**Screening** |
| Day 2 cont. | Immunomagnetic Bead Capture & Rainbow Agar Plating |  
| Day 3 | O157 Latex Agglutination & Sheep Blood Agar Plating | presumptive (+)  
**Confirmation** |
| Day 4 | Examine for Control Bioluminescence & H7 Agglutination |  
| Day 4 cont. | ELISA Shiga Toxin Assay |  
| Day 4 cont. | Biochemical Identification | confirm (-,+)  
|
Non-O157 STEC Program

- STEC = “Shigatoxigenic *E. coli*”
  - Six Non-O157 serotypes are targeted
    - O26, O111, O103, O45, O121, O145
  - Serotype strain must have *stx* (Shiga toxin) and *eae* (intimin) genes
  - Currently FSIS only analyzes beef manufacturing trimmings for Non-O157 STECs
Larger *E. coli O157:H7* and Non-*O157* STEC Test Portions

- Larger test portions (325-375 grams) are most important for “n60” and other composite samples containing many samples.
- Less important for single “grab” samples of ground beef final product testing when:
  - Trim and components have already been tested using robust sampling and 325-375-gram test portions
  - Multiple samples are collected throughout the production day
- Methods must be adapted, optimized and validated for effective use with 325-375 gram test portions
Food Safety and Inspection Service:

**E. coli O157:H7 and Non-O157 STEC Testing Concerns**

- Supplemental validation and special instructions for testing larger test portions
  - For enrichment periods <15 hours
  - 325-375g test portions typically often require longer minimum enrichment period than 25g

- Culture-based detection and confirmation requires immunomagnetic separation (IMS)
Food Safety and Inspection Service:

**Listeria Testing**

- Includes:
  - *L. monocytogenes* testing (FSIS)
  - *Listeria*-like or *Listeria* spp. testing (industry)
Food Safety and Inspection Service:

**Listeria monocytogenes (MLG Ch. 8)**

<table>
<thead>
<tr>
<th>Day</th>
<th>Task Description</th>
<th>Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 1</td>
<td>Sample Prep and Primary Enrichment&lt;br&gt;Stomach 25g sample + 225 ml LEB&lt;br&gt;Incubate 20-24 hrs @ 30°C</td>
<td>Confirm (-)</td>
</tr>
<tr>
<td>Day 2</td>
<td>Secondary Enrichment&lt;br&gt;Inoculate 0.1ml to MOPS-BLEB&lt;br&gt;Incubate 18-24hrs at 35°C</td>
<td>Screening</td>
</tr>
<tr>
<td>Day 3</td>
<td>Perform BAX PCR&lt;br&gt;Streak all PCR positive samples to MOX&lt;br&gt;Incubate 24-28hrs at 35°C</td>
<td>Confirm (-)</td>
</tr>
<tr>
<td>Day 4</td>
<td>Pick Typical Colonies&lt;br&gt;Pick 20 colonies and collectively streak for isolation on HBO&lt;br&gt;Incubate 18-26hrs at 35°C</td>
<td>Confirmation</td>
</tr>
<tr>
<td>Day 5</td>
<td>Streak isolated colony to HBO</td>
<td>Presumptive (+)</td>
</tr>
<tr>
<td>Day 6</td>
<td>Perform biochemical testing and Inoculate CAMP test&lt;br&gt;Streak HBO plate</td>
<td></td>
</tr>
<tr>
<td>Day 7</td>
<td>Perform Ribosomal RNA based testing</td>
<td>Confirm (-/+)</td>
</tr>
</tbody>
</table>
Food Safety and Inspection Service:

**Expectations for *Listeria* Environmental Testing Equivalence**

- Compliance Guidelines, May 2006, pp. 42-44
- For optimal sensitivity of detection, method for food contact surface testing must:
  - Validated by a recognized body (e.g., AOAC, AFNOR)
  - Be enrichment-based
  - Enrich the entire sponge/swab sample
    - For example an aliquot from sponge/swab does not provide opportunity to detect bacteria trapped in the sponge
Food Safety and Inspection Service:

Analytes for Industry Food Contact or Environmental Surface Testing

- Establishment laboratories test for one of the following:
  - *Listeria monocytogenes*: Use internationally recognized enrichment-based method that biochemically confirms culture as *L. monocytogenes*
  - *Listeria* spp.: Use internationally recognized enrichment-based method that uses ELISA, PCR or other screening technology to provide more rapid but less specific *Listeria* spp. result
  - “*Listeria*-like” indicator bacteria: Use the first part of an internationally recognized enrichment-based method to find suspect *Listeria* colonies (e.g., darkened colonies on MOX using the FSIS method)
Salmonella Testing

• Raw products
  – Meat and turkey carcass sponge samples
  – Chicken carcass/parts rinsates
  – Raw meat and comminuted poultry

• Processed products
  – RTE (325g portion)
  – Pasteurized egg
Food Safety and Inspection Service:

**Salmonella (MLG Ch. 4)**

<table>
<thead>
<tr>
<th>Day 1</th>
<th>Sample Prep and Primary Enrichment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Stomach sample + BPW</td>
</tr>
<tr>
<td></td>
<td>Incubate 20-24 hrs at 35°C</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Day 2</th>
<th>Perform PCR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>All samples that do not test PCR negative are carried forward to RV and TT broth</td>
</tr>
<tr>
<td></td>
<td>Incubate 22-24 hrs at 42°C</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Day 3</th>
<th>Streak RV and TT on BGS and DMLIA plates</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Incubate 18-24 hrs at 35°C</td>
</tr>
</tbody>
</table>

| Day 4 | Pick suspect colony from Plate medium to TSI and LIA slants. Incubate slants with loosened caps for 22-24 hrs at 35°C |

<table>
<thead>
<tr>
<th>Day 5</th>
<th>Perform O and H serology on slants. Streak on SBA for biochemical testing</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Incubate 18-24 hrs at 35°C</td>
</tr>
</tbody>
</table>

| Day 6 | Perform biochemical testing using colony from SBA plate. |

**Enrichment**

**Screening**

**Confirmation**
Campylobacter Testing

- Qualitative or quantitative
  - Semi-quantitative for regulatory application

- Target = *C. jejuni*, *C. lari* or *C. coli*
# Campylobacter (MLG Ch. 41) - Quantitative

<table>
<thead>
<tr>
<th>Day 1-2</th>
<th>Direct Plating (no enrichment)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Direct plating onto Campy-Cefex</td>
</tr>
<tr>
<td></td>
<td>Incubate 48 hrs at 42°C</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Day 3</th>
<th>Confirmation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Count colonies</td>
</tr>
<tr>
<td></td>
<td>Pick 5 typical colonies</td>
</tr>
<tr>
<td></td>
<td>confirm (-)</td>
</tr>
<tr>
<td></td>
<td>Microscope examination</td>
</tr>
<tr>
<td></td>
<td>for morphology/motility</td>
</tr>
<tr>
<td></td>
<td>confirm (-/+ )</td>
</tr>
<tr>
<td></td>
<td>Latex agglutination</td>
</tr>
</tbody>
</table>

**RESULTS ARE REPORTED AS POSITIVE/NEGATIVE**

≥ 1 CFU = POSITIVE
### Campylobacter (MLG Ch. 41) - Qualitative

#### Sample Prep and Primary Enrichment
- Stomach sample + BF-BEB
- Incubate 48 hrs at 42°C

#### Enrichment

#### Plating/isolation
- Incubate 48 hrs at 42°C

#### Plating/isolation onto Campy-Cefex

#### Day 5
- Count colonies
- Pick 5 typical colonies
- Microscope examination for morphology/motility
- Latex agglutination
- confirm (-) **Confirmation**
- confirm (-/+)

**RESULTS ARE NOT USED FOR REGULATORY PURPOSES**
Food Safety and Inspection Service:

Issues for *Campylobacter* Testing

- *Campylobacter* is highly vulnerable to freezing
  - Do not freeze samples
- Can be a challenging test (inconsistent results across labs)
Quantitative Testing

• NOTE: Quantitative testing typically cannot accommodate larger test portions and provide the opportunity for detection that a qualitative test can provide

• Two options:
  – MPN
  – Direct plating
Most Probable Number (MPN) Enumeration Analysis

- Traditional enrichment-based analyses are performed on three or more dilutions, each typically in triplicate, from a single sample homogenate \((i.e., \text{MPN} = \text{method format, not a specific method per se})\)
- **Advantages:**
  - Better sensitivity (lower LOD) than direct plating
- **Disadvantages:**
  - Very resource intensive/expensive
  - Test portion \(\leq 3.3\) grams (FSIS method = \(< 33\) grams)
- **Application:**
  - For quantifying low levels of pathogens \((e.g., \text{Salmonella, E. coli O157:H7, L. monocytogenes})\)
Food Safety and Inspection Service:

Quantitative Testing - MPN (most probable number)

325 grams + 10 fold buffer = 0.1 grams/mL

Dilute 1:10, 1:100

10 mL (1 gram x 3)

10 mL 1:10
(0.1 gram x 3)

10 mL 1:100
(0.01 gram x 3)

enrich

+++  

++  

--
Quantitative Testing - MPN (most probable number)

Example:

“3-2-1” = Y MPN/g
(use MPN table to determine Y)

Total tested:
3.33 grams
(33 grams FSIS method)

Level of Detection =
< 0.3 MPN/gram (0-0-0)
<0.03 MPN/gram (FSIS method)
Direct Plating Enumeration Methods

- Product is homogenized in diluent and small volume is directly dispensed onto agar media (i.e., sometimes there is a 1-2 h “resuscitation” step, but *enrichment* is never used prior to plating)

- Advantages:
  - Allows easy inexpensive quantitative analysis

- Disadvantages:
  - Accommodates only a very small test portion
  - Higher LOD (i.e., often 100 CFU/g) not suitable for detecting low levels of pathogens

- Application:
  - Expedient for higher level analytes (e.g., indicators, *Campylobacter*, *S. aureus*, *C. perfringens*, *B. cereus*)
Food Safety and Inspection Service:

**Quantitative Testing: Direct Plating**

325 grams + 10-fold buffer = 0.1 grams/mL

1 mL (0.1 gram) Dilute 1:10, 1:100

No enrichment

6 cfu/1 mL/0.1 g = 60 cfu/g

1 mL 1:10 (0.01 gram)

1 mL 1:100 (0.001 gram)

Total tested 0.11 grams

Level of Detection = <10 cfu/gram (0 cfu from homogenate)
Food Safety and Inspection Service:

The Establishment and the Laboratory
Food Safety and Inspection Service:

**Establishment and Laboratory Communication is Vital**

- The communication challenge
  - The establishment may not understand the testing conducted on their behalf
  - The laboratory does not necessarily know what the establishment needs
  - The laboratory may not be aware of special validated procedures for larger test portions
- The establishment is ultimately responsible
Establishment Documentation

- Does the establishment have the necessary documentation?
  - Can the establishment provide the method used for microbial detection?
  - Can the establishment provide evidence that the method used was properly validated by an independent body?
  - Can the establishment explain why the method fits the need?
Food Safety and Inspection Service:

**Issues for Industry Labs**

- On-site vs. off-site labs
  - Shipment of samples/handling during shipment
- Overarching concerns for on-site labs
  - Is testing effective?
  - Is testing safe in that facility?
  - Enrichment of pathogens in an establishments
- Evaluate the following:
  - Are personnel qualified?
  - Does the lab have proper equipment and materials for testing and disposal of contaminated media?
  - Do they follow the validated testing protocol?
ISO 17025 Laboratory Accreditation

• ISO 17025 = protocol for establishing and documenting a microbiology laboratory quality program (i.e., “HACCP” for labs)

• Accrediting bodies = A2LA and others

• Accreditation implies robust quality program but does not necessarily indicate methods meet FSIS expectations
Food Safety and Inspection Service:

Helpful Guidance
Food Safety and Inspection Service:

**Existing Agency Guidance – Compliance Guides**

- DRAFT FSIS Compliance Guideline for Controlling *Salmonella* and *Campylobacter* in Raw Poultry (Fourth Edition, December 2015)
- Compliance Guideline for Sampling Beef Trimmings for Shiga Toxin-Producing *Escherichia coli* (STEC) Organisms or Virulence Markers (August 2014)
- Establishment Guidance for the Selection of a Commercial or Private Microbiological Testing Laboratory (June 2013)
- FSIS Guidance for Evaluating Test Kit Performance (October 2010)
- Enforcement, Investigations, and Analysis Officer (EIAO) Food Safety Assessment (FSA) Methodology (FSIS Directive 5100.1, Rev. 4)
Existing Agency Guidance - askFSIS

• askFSIS Q&A sometimes contains additional information on testing methods
• If you cannot answer your question there, please submit to askFSIS
Questions?
Ask me now...

or for future questions:
• Enter question into askFSIS
• Provide documentation for review
• Request “Sampling Queue”