Approval of Food Microbiological Methods in Canada

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Outline

- Food Safety in Canada
- The Microbiological Methods Committee (MMC) and the *Compendium of Analytical Methods*
- MMC’s Method Validation Guidelines and Submission Evaluation Process
- MMC Method Submission Packages – Areas for Improvement
Food Safety in Canada
Food Safety in Canada

• Food offered for sale in Canada is required to be manufactured, stored, transported and otherwise handled under conditions that provide for its microbiological safety and general cleanliness.

• Legislative framework is mainly based on the *Food and Drugs Act*.

• Other legislation includes:
  - *Canada Agricultural Products Act*
  - *Meat Inspection Act*, etc.
  - *Safe Food for Canadians Act*

• A joint responsibility of government, industry, and consumers.
Food Safety Responsibility

Health Canada (HC):

- develops food safety standards and policies to help minimize the risk of foodborne illnesses

The Public Health Agency of Canada (PHAC):

- conducts outbreak surveillance and epidemiology and provides advice to protect people’s health

The Canadian Food Inspection Agency (CFIA):

- carries out inspection of the food industry to ensure that it meets its food safety responsibilities
Food Safety Responsibility and Microbiological Testing

• Food producers are responsible for ensuring the safety of their products:
  ➢ Design, development and implementation of effective preventive food safety control systems

• Microbiological testing can be used as one of a series of tools used in a food safety system

• Microbiological food testing is carried out by the Government of Canada to support food safety responsibilities

No sampling plan or end product testing can guarantee food safety or quality.
Use of Microbiological Methods in the Food Industry

• CFIA-mandated testing: **use a method in the Compendium**
  - Meat Hygiene Manual of Procedures, under the *Meat Inspection Act* and *Meat Inspection Regulations*
    - Test precursor materials used for the production of finished raw ground beef products for *E. coli* O157:H7
    - Plants producing Ready-to-Eat (RTE) meat and poultry products require an environmental sampling program for testing food contact surfaces for *Listeria* spp.

• Testing for other purposes: **use any fit for purpose method**
  - Trend monitoring
  - HACCP verification
  - Quality control of ingredients
A.01.012. The Director shall, upon request, indicate that a method is acceptable or otherwise upon its submission to him for a ruling.

A.01.010. In these Regulations, “acceptable method” means a method of analysis or examination designated by the Director as acceptable for use in the administration of the Act and these Regulations.

“Director” means the Assistant Deputy Minister, Health Products and Food Branch, of the Department of Health.
The Microbiological Methods Committee (MMC) and the Compendium of Analytical Methods
The Microbiological Methods Committee (MMC)

Mandate:
• To provide methodology to support Health Canada and Canadian Food Inspection Agency mandates in overseeing the safety of the Canadian food supply

Objective:
• To ensure that methodology:
  ➢ Is reviewed in a timely, objective and transparent manner
  ➢ Has undergone appropriate validation and testing for ruggedness
  ➢ Is based on sound science
  ➢ Is fit for purpose
The Microbiological Methods Committee (MMC)

• All method submission packages assessed by the MMC are treated as **confidential**

• Strive to ensure that transparency is upheld, while respecting the duty to confidentiality

• Additional stakeholders include:
  - Private laboratories
  - Academia
  - Industry
  - International laboratories
The *Compendium of Analytical Methods*

A ready reference of the methods used by Health Canada, CFIA, and other agencies and organizations:

- To determine compliance of the food industry with standards and guidelines
- To assess the microbial quality of foods
- In support of foodborne disease investigation
- In support of Health Canada policy development
- In food plant environment and HACCP testing
The *Compendium of Analytical Methods*

**Volume 1:** Official Methods - Microbiology, Supporting Documents

**Volume 2:** HPB Methods - Microbiology

**Volume 3:** Laboratory Procedures - Microbiology

**Volume 4:** Extraneous Material

**Volume 5:** Parasites, Viruses, and other Foodborne Pathogens
MMC’s Method Validation Guidelines and Submission Evaluation Process
Introduction to Method Validation

- Repeated use of a method

Method Validation

- The provision of objective evidence that a method fulfills specified requirements
- The process of establishing the performance characteristics and limitations of a method
MMC Guidelines

- Recall that MMC mandate includes regulatory obligation in overseeing food safety
- Relative validation: evaluate performance parameters of a method in comparison to an accepted cultural reference method
- Part 4: Guidelines for the relative validation of indirect qualitative food microbiological methods

General Overview - Method Submission Package

• Pre-collaborative study:
  - comparison to an accepted cultural reference method
• Inclusivity / exclusivity studies
• Limit of detection study
  - the smallest number of culturable microorganisms detectable in 50% of the samples
  - Annex 4.5, *Determination of the Limit of Detection*
• Transfer study
  - to demonstrate the new method’s performance in another laboratory, under the control of the submitting laboratory
Relative Validation

• Results of the alternative method are compared to that of the cultural reference method

• Statistical tools and performance criteria are used for assessment of results

• Paired samples test the same analytical portion by both methods using portions of one common enrichment broth
  ➢ Confirmation is simplified

• Unpaired samples test two different analytical portions with two different enrichment broths
  ➢ Confirmation is more laborious
Confirmation for Paired Samples

Reference Method

Alternative Method

Reference (R)  
Reference result serves as confirmation

Alternative Presumptive (AP)

Alternative Final (AF)
Confirmation for Unpaired Samples

Reference Method

Divert to the reference method

Cultural confirm. of alternative enrichment

Reference (R)

Alternative Presumptive (AP)

Alternative Final (AF)

Alternative Method
Why Divert to the Reference Method?

Duration of enrichment: 96 hours

Duration of enrichment: 24 hours

Reference Method

Alternative Method

Alternative Presumptive (AP)

Alternative Final (AF)

2 analytical portions = different true status possible

Is the confirmation procedure adequate?

Would the results change if the incubation was longer?
Performance Criteria

All methods published in the *Compendium of Analytical Methods* must meet or exceed the following criteria:

1. Sensitivity $\geq 98\%$ (ability to detect target)
2. Specificity $\geq 90.4\%$ (response for target only)
3. False Negative Rate $< 2\%$
4. False Positive Rate $< 9.6\%$
5. Efficacy $\geq 94\%$ (degree of correspondence)
6. Lower Limit of Detection must be comparable to the standard method (usually 3-5 CFU/25 g) or must detect $\leq 3$ CFU/g
Help with Method Validation

• Compendium of Analytical Methods, Volume 1, Development of Methods
  

• Contact the Microbiological Methods Committee:
  mmc-cmm@hc-sc.gc.ca

• Consult with laboratories accredited by Palcan (Standards Council of Canada) for “Test Method Development and Evaluation and Non-routine Testing”
  
  http://palcan.scc.ca/SpecsSearch/TLSearchForm.do
Current international interest in determining the equivalence of international reference methods

- Variation in cultural methods for *Listeria monocytogenes* between jurisdictions
- *E. coli* O157 as jurisdictions adjust enrichment conditions to favour all VTEC (e.g., lowered novobiocin concentration)

Choice of cultural reference methods is important

If possible, relative validation against a Canadian cultural reference method is recommended

Current international interest in harmonization of method validation guidelines

Participation in ISO TC34/SC9 “Food Microbiology”
MMC Submission Evaluation Process

Method
Submission Package received

Submission Package checked for completeness

Technical Group (TG) established

Submission Package incomplete

Communicate with submitter
MMC Submission Evaluation Process

TG assesses method validation package against MMC criteria

TG identifies additional information needed

MMC relays information between submitter and TG

- TG will request information until recommendation can be made
- Submission packages with missing or unclear information may not be identified until this stage
MMC Submission Evaluation Process

- MMC SC reviews TG recommendation
- BMH Director makes final decision
- MMC notifies submitter of decision
MMC Submission Evaluation Process

Submitter provides written MFLP method

MMC prepares the method for publication in the *Compendium of Analytical Methods*

Compendium subscribers notified by e-mail

- Review against validation package
- Translate and verify
- Format for web publication
- Internal MMC approval

MMC Method Submission Packages – Areas for Improvement
Study Components that are Usually Satisfactory

- Inclusivity/Exclusivity Study
- Number of Samples (20 low/fractional, 20 high, 5 uninoculated)
- 3 food types validated per food category
- 5 relevant spiking strains used
- Paired/unpaired samples identified
- Raw and summary data provided
- LOD study
Areas for Improvement

• Completion of Part 2B template
  • Fill out in detail (do not provide yes/no answers)

• Choice of cultural reference method
  • International interest in the equivalence of international reference methods
  • *L. monocytogenes*, *E. coli* O157:H7

• Confirm all positive and negative alternative method results

• Either the alternative or reference method must yield fractional positives
  • 5 – 15 positives / 20 samples (25 – 75%)

• Use an appropriate equilibration period
Areas for Improvement

Interference Organisms

• Particularly with *L. monocytogenes* / *L. innocua*
• Generic *E. coli* and non-O157 VTEC
• Interference organisms should be present at approximately 10 times the target organism
• Test one food type per food category for interference organisms
Areas for Improvement

Selection of Foods

- Foods chosen should:
  - Use a variety of food items to represent variety within the food type AND
  - Be a relevant risk for the target, or a difficult matrix for the target

Target Organism: *E. coli* O157:H7

<table>
<thead>
<tr>
<th></th>
<th>Apple Juice (shelf stable tetra pack)</th>
<th>Unpasteurized Apple Cider</th>
</tr>
</thead>
<tbody>
<tr>
<td>Commercially sterile</td>
<td>Complex microbiota</td>
<td></td>
</tr>
<tr>
<td>Irrelevant risk</td>
<td>Relevant risk</td>
<td></td>
</tr>
</tbody>
</table>

Not a good choice for validation

An excellent choice for validation
Quantify the level of stress in the inoculum

- Recommended range of 50 – 80%
- Percent injured cells = 100 - ([sel. count/non-sel. count] x 100)
- Example: A culture of *L. monocytogenes* is heated at 50°C for 10 min. Plating results are $10^4$ cfu/ml on MOX and $10^5$ cfu/ml on TSA.

\[
\text{% injured} = 100 - ([10^4 / 10^5] \times 100)
\]

\[
= 90
\]

- 90% stress is equivalent to a 1 log$_{10}$ decrease on selective agar, relative to the count on non-selective agar
- $10^4$ cfu/ml are not stressed/injured and there are $9 \times 10^4$ cfu/ml that are stressed/injured
Thank You