



Canadian Food
Inspection Agency

Agence canadienne
d'inspection des aliments

Canadian Food Inspection Agency



Our vision:

To excel as a science-based regulator, trusted and respected by Canadians and the international community.

Our mission:

Dedicated to safeguarding food, animals and plants, which enhances the health and well-being of Canada's people, environment and economy.

Implementation of genomics technologies in regulatory food microbiology testing

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Canadian Food Inspection Agency

Canada

Workshop on the application of genomics tools for the rapid molecular characterization of bacterial isolates in foodborne disease outbreak investigations (Ottawa, February 24&25, 2014)

Audience

- Federal/Provincial and International food safety regulatory and research experts

Objectives

- Overview of genomics in food safety regulation
 - Current state – characterization of microbial hazards & regulatory decision making
 - Current tools for detection, typing and risk profiling
 - New possibilities for genomics
- Gap analysis

Outcomes

- Better understanding of how to integrate genomics in decision processes
- Identification of research gaps
 - Method development
 - Regulatory decision making
 - Regulatory policy

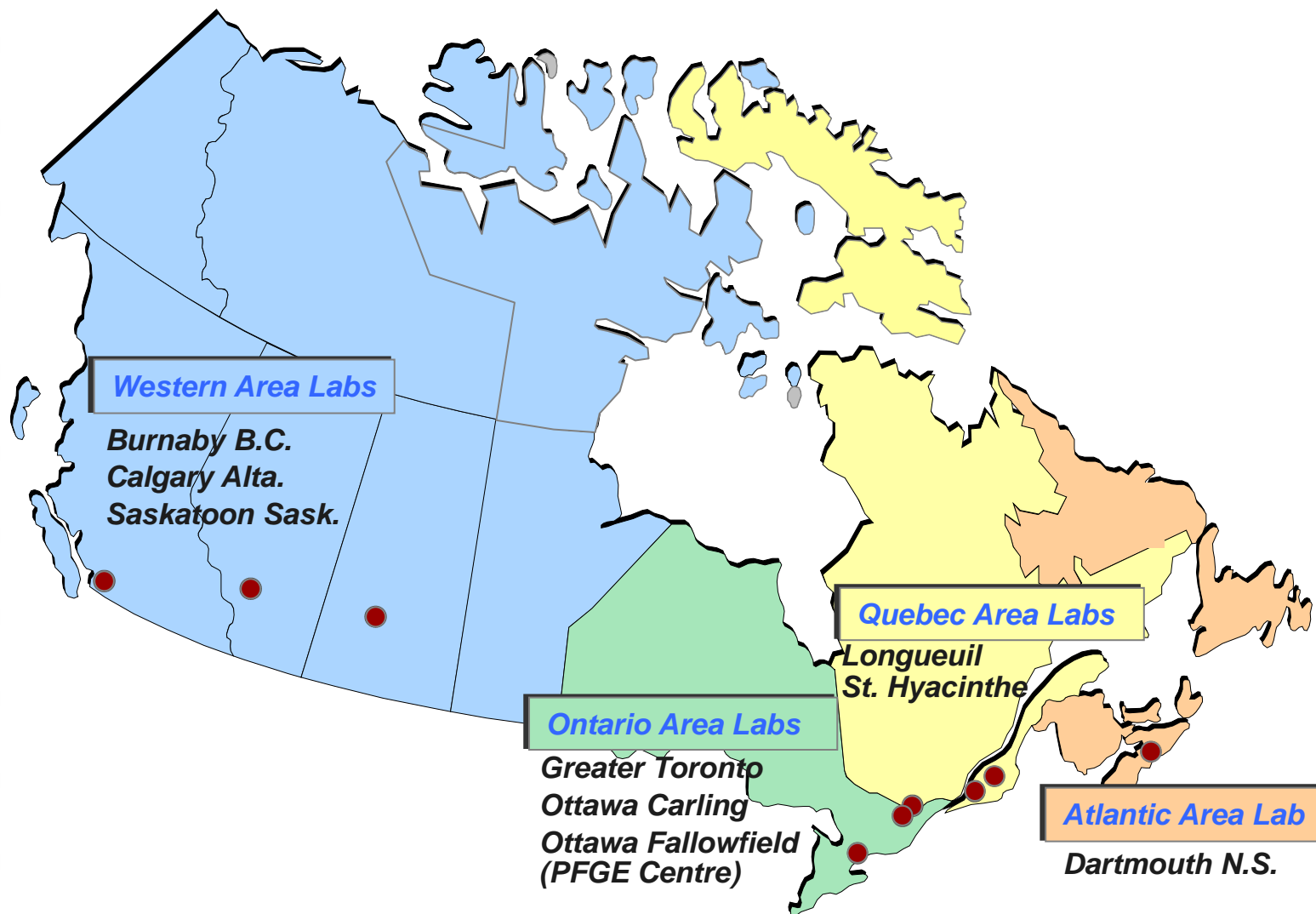
Who We Are

The CFIA is...

- Canada's largest science-based regulatory agency
- responsible for the delivery of all federally-mandated programs for food inspection, plant and animal health, and consumer protection as it relates to food
- mandated to safeguard Canada's food supply and the plants and animals upon which safe and high-quality food depends
- *Key objective to protect Canadians from preventable health risks, by verifying compliance of the food industry with applicable standards through comprehensive inspection and testing activities (safety, nutrition, authenticity & composition)*



Food Laboratories



Food Microbiology Testing

Routine Monitoring

- Indicator organisms: Generic E. coli, coliforms, yeast, mold...
- Food-borne pathogens (bacterial, viral, parasitic)
- Safety parameters – Salt content, pH, Aw
- Standards established by Health Canada, International Organizations (e.g. *Codex Alimentarius*)
- Food Programs:
 - Meats, Fish, Dairy & Egg products, honey
 - Fresh & Processed Fruits and Vegetables
 - Manufactured Foods (bread, infant formula, spices...)
 - Food processing environment samples

Food Microbiology Testing

Targeted surveys (FSAP)

- 2010: Tomatoes → *Salmonella*/*Shigella*/*E.coli* (0/1414)
- 2010: Produce → *Salmonella* (2/4250), *Listeria* (7/1850)
- 2011: Cantaloupe → *Salmonella* (1/499)



Consumer complaints

- Restaurants, food services, retail conditions
- Food quality, safety, labelling

Outbreak investigation

- Source attribution
- Scope of contamination
- Hazard mitigation
- PFGE (PulseNet)



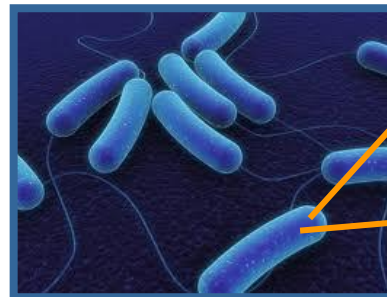
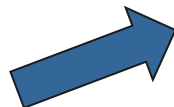
Role of genomics in CFIA food testing programs

WGS of food-borne pathogens

- Identification (definitive proof of presence)
- Virulence profiling (armed and dangerous)
- Sub-typing (timely outbreak recognition & traceback, source tracking)

Method development

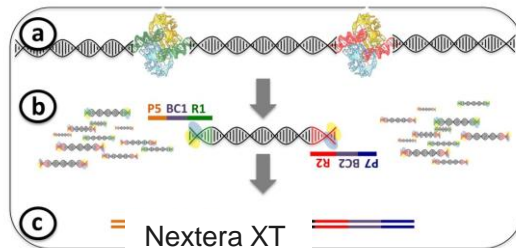
- *in silico* validation
- *ad hoc* methods



ATCCGTACGTA
AAGGGCCTAG
CTTGGACTTTG
GGGATCGCTA

Whole Genome Sequencing

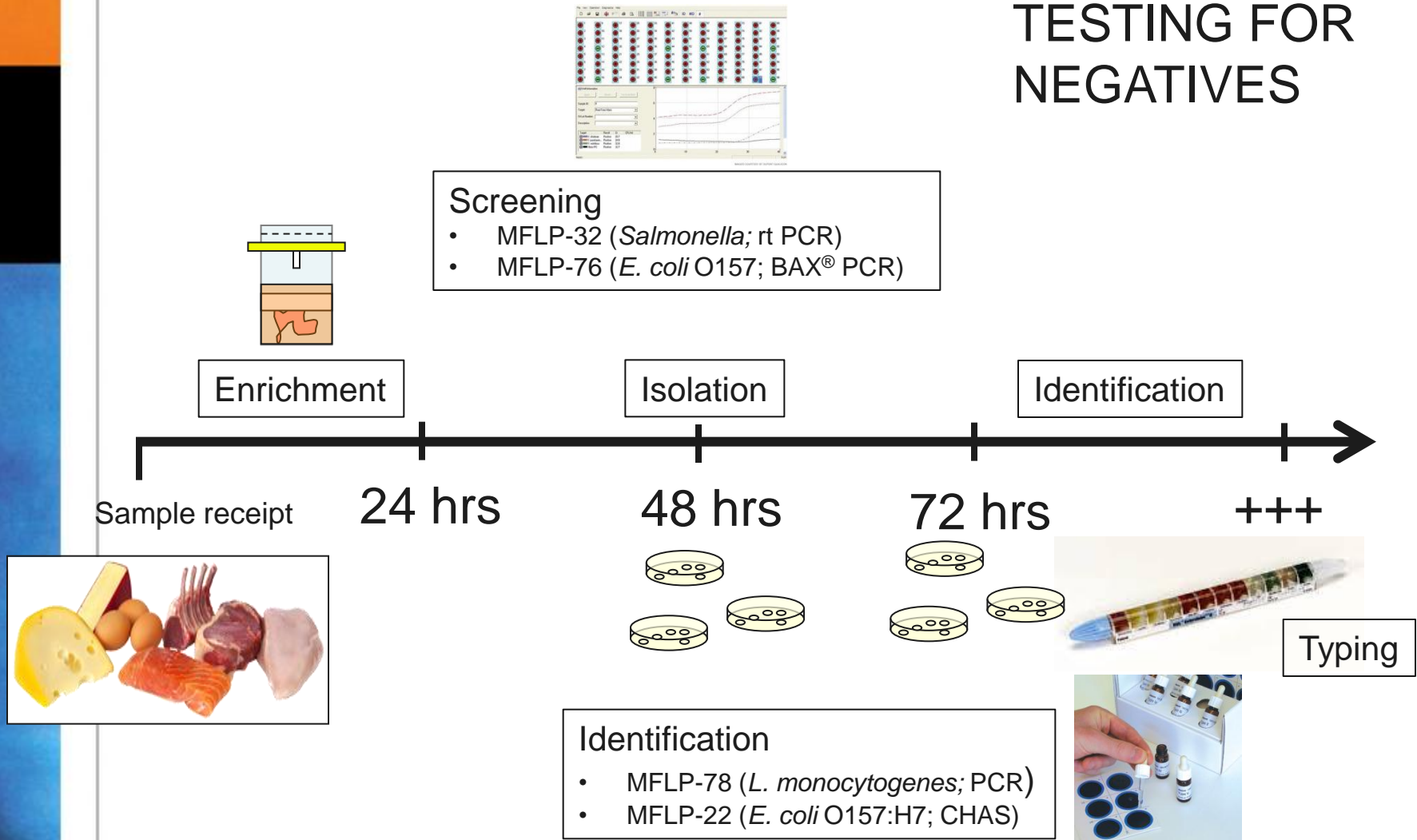
- All strains collected through food testing programs
- Historical collections (*Salmonella*, *E. coli*, *Listeria monocytogenes*)
- Deployment of capacity to front line testing laboratories?



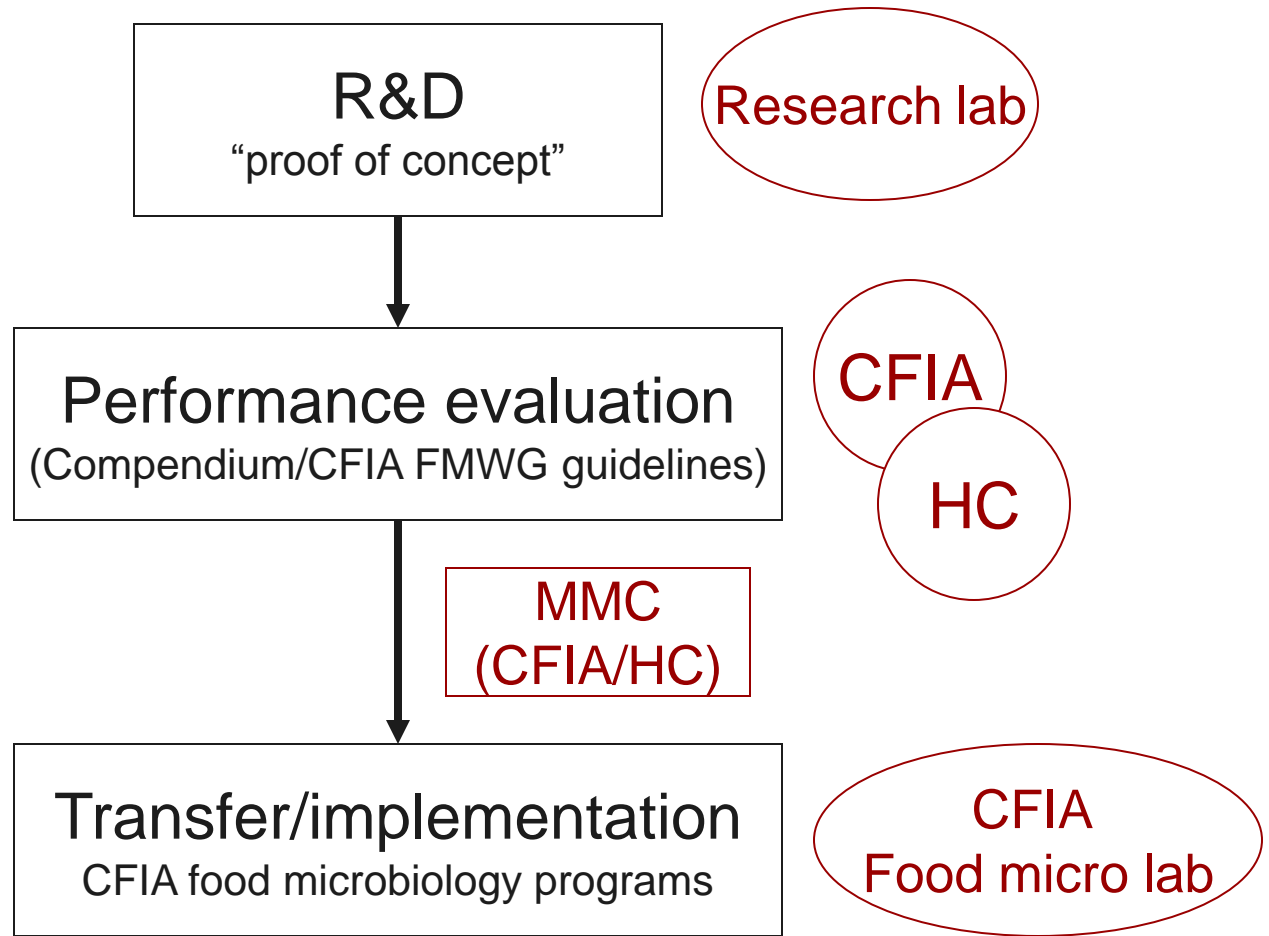
MiSeq

Regulatory food testing

TESTING FOR NEGATIVES



Development and implementation of food microbiology test method



Validation of genetic targets for bacterial identification

PCR method validation:

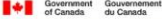
- Inclusivity: 75-150 target strains
- Exclusivity: 50 strains
- Recommendations for selection of strains
- Performance criteria:
 - Relative Sensitivity $\geq 98\%$,
 - Relative Specificity $\geq 90.4\%$,
 - False negative rate $< 2.0\%$,
 - False positive rate $< 9.6\%$,
 - Efficacy $\geq 94\%$




In silico validation of PCR targets

Virtual strain bank:

- Inclusivity: >1000 genetically diverse target strains
- Exclusivity: >5000 strains
- Food specific-metagenome
- Standardization of validation process

 **Welcome / Bienvenue**

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Virtual Primer Validation

Left primer:

Right primer:

Mismatches per primer <=

Display alignments:

Target Organism:

PCR Conditions:

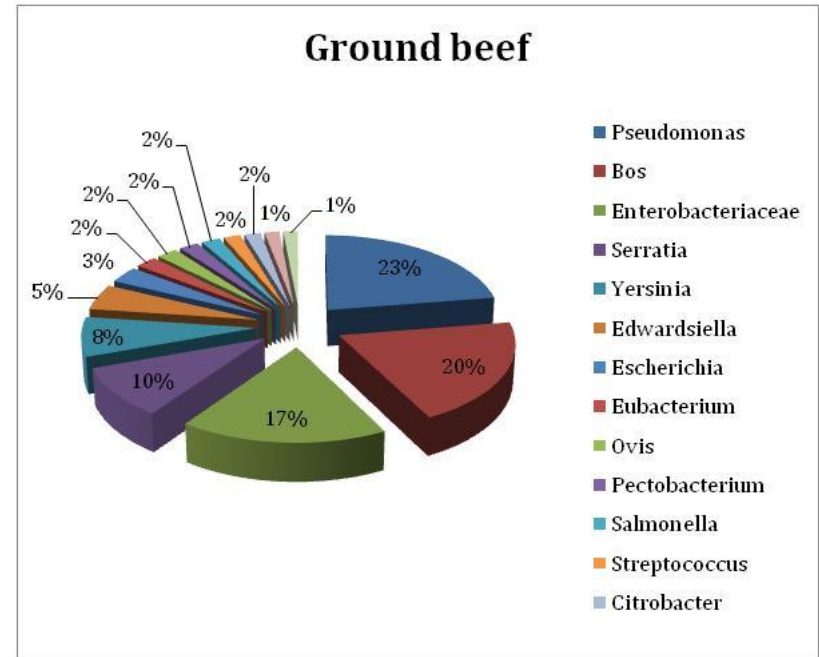
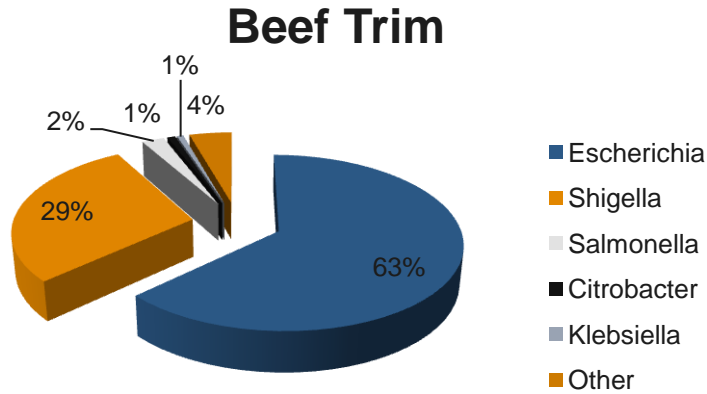
Target Organism: Escherichia_GB072013

Left primer: CCGATACGCTGCCAATCAGT Ta= 75.22 C
Right primer: ACGCAGACCGTAGGCCAGAT Ta= 77.27 C

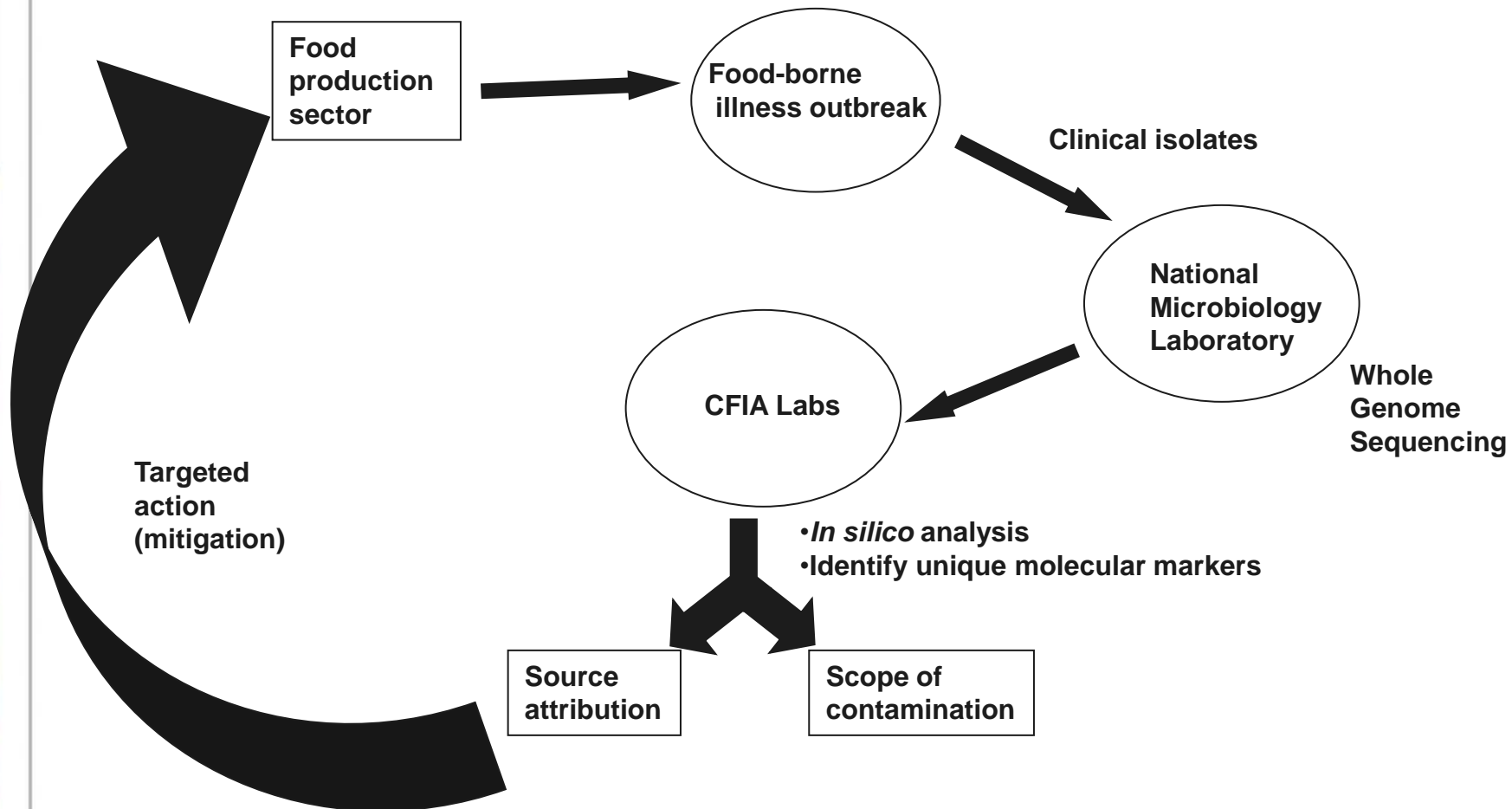
Inclusivity:
Targets are amplified in 58 of 60 target genome sequences (96.67%)

Exclusivity:
Targets are amplified in 0 of 268 non-target genome sequences (0.00%)

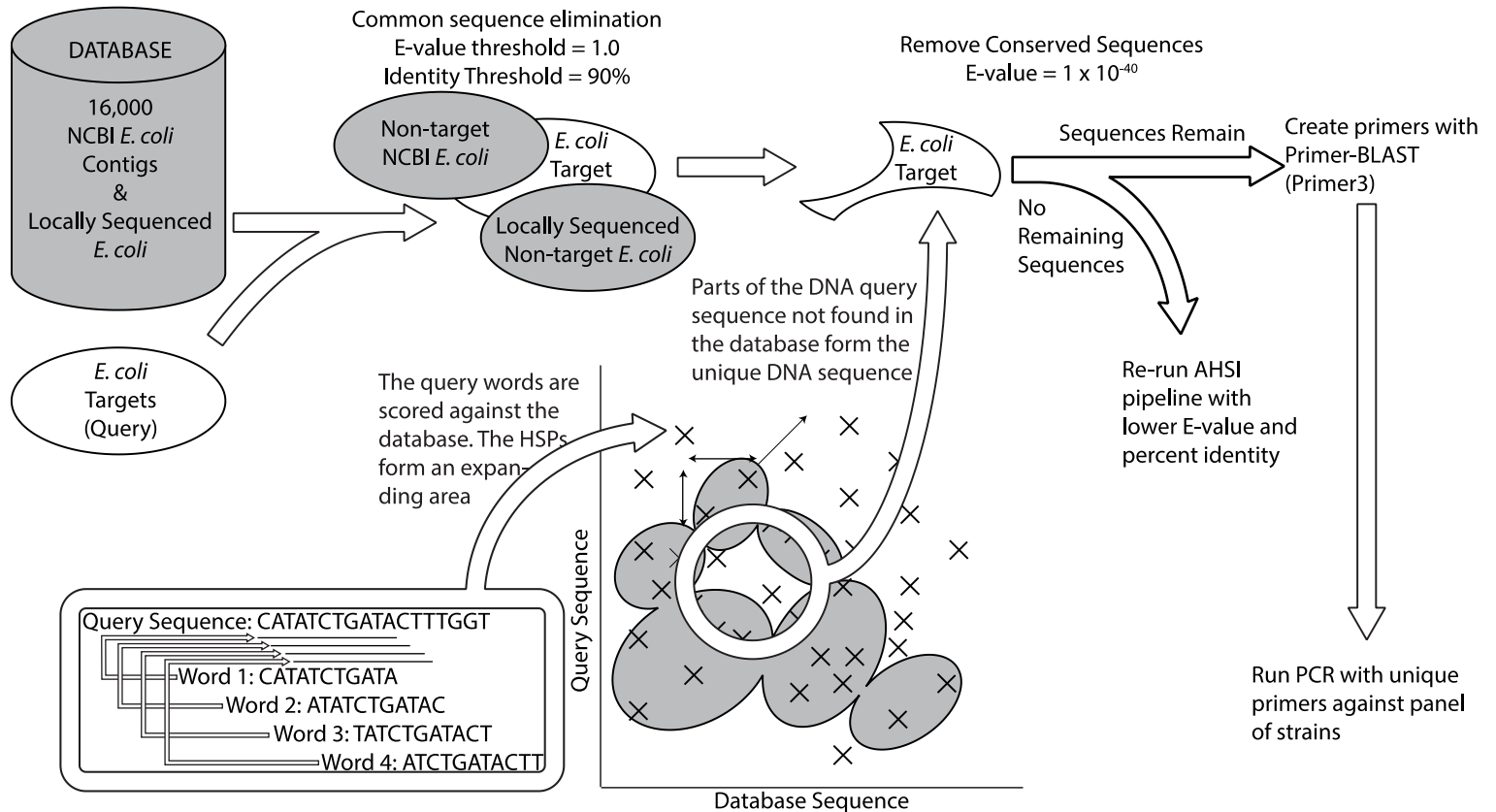
Meat metagenome database to support *in silico* PCR validation



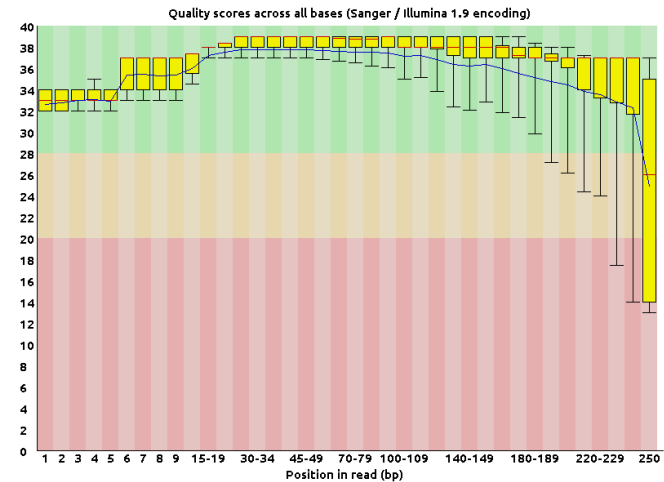
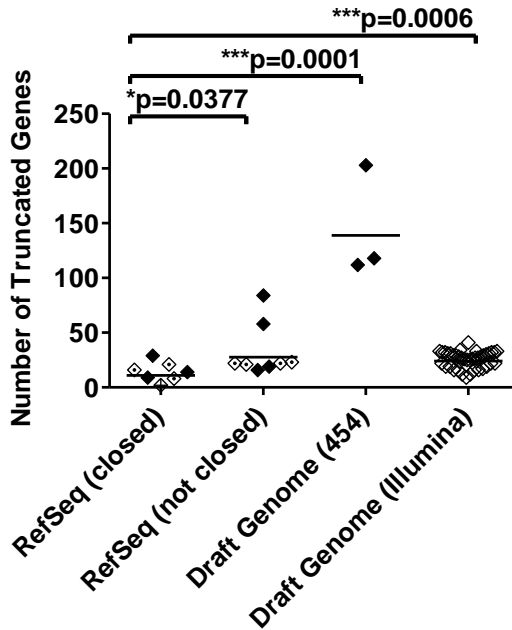
Deployment of *ad hoc* methods in support of outbreak investigations



Identification of unique genome markers



Quality control



CFIA has been accredited to ISO/IEC 17025 **General requirements for the competence of testing and calibration laboratories** since 1997 and to CAN-P-1595: **Requirements for Accreditation of Laboratories Engaged in Test Method Development & Non-Routine Testing** (2005).

Global Coalition for Regulatory Science Research Bioinformatics working group:

The establishment of "best practices" for the analysis and reporting of NGS data for regulatory science
 ensure reliability, consistency and auditability of processes used in the production of genomic information

Acknowledgement

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and Security Program



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