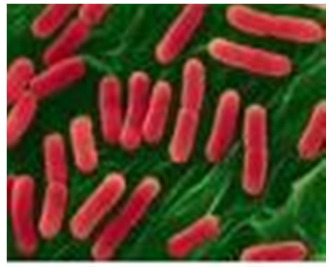
INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Testing for Non-O157:H7 Shiga Toxin-Producing Escherichia coli Serotypes in Ohio

Jodi Taylor
IFPTI 2011 Fellow
Ohio Department of Agriculture
Division of Meat Inspection

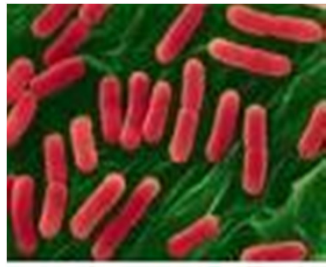
Background

- *E. coli* O157:H7 most common strain associated with foodborne illness in the United States
- FSIS slated to include 6 additional serotypes as adulterants in June 2012
  - Known as the "Big 6"
  - O103, O111, O26, O45, O121, O145
- The "Big 6" are not routinely tested for
  - Difficulty in identifying and confirming
  - Lack of validated laboratory methods



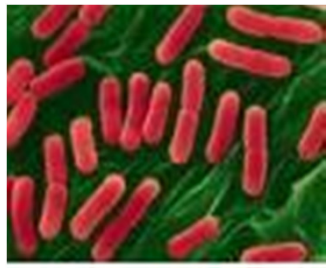
Background (continued)

- Ruminant animals are primary transmission source
  - Food products (beef products)
  - Manure-contaminated vegetables
  - Animal to human contact
- Ground beef is being targeted for this research



Background (continued)

- The Ohio Meat Inspection program only analyzes beef for *E. coli* O157:H7
  - Ground beef sampling program started in 2002
  - Trim sampling started in 2006
  - Over 12,000 samples analyzed since 2008
  - Seven confirmed positive findings (4 in 2009; 3 in 2011)



INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Problem Statement

- DMI has no baseline for the ‘Big 6’ serotypes.
- Limited test kits commercially available.
- Labs have no experience screening for non-O157:H7.
- Validation of screen kits is a concern.

Research Questions

1. Have the analytical methods been validated enough to produce accurate results?
2. Is the ODA DMI ready to regulate for these additional E. coli serotypes?
3. Is it possible to determine if non-O157:H7 STEC is present in ODA DMI-inspected beef slaughter facilities?

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Methodology

- All of Ohio's beef slaughter operations had potential for being included in project.
- 100 samples were analyzed for both *E. coli O157:H7* and non-O157:H7.
- Inspectors responsible for random collection of samples and establishments sampled were briefed on research details.
- Samples were collected from August 22, through September 29, 2011.

Methodology (continued)

- Blind samples were used for the non-O157:H7 portion of the study.
  - Facilities were identified for the *E. coli O157:H7* portion.
- Samples were analyzed within 24 hours of collection.
- Analysis was conducted by the ODA Consumer Protection Laboratory (CPL).

Methodology (continued)

- Samples were screened using two methods
  - Bio-GX
    - Reported as potential positive if confirmed for the stx1 or stx2 gene
    - Potential positives were further analyzed using the kit's two-plate system to identify serotype

Methodology (continued)

- DuPont BAX
  - 60 samples analyzed
  - Screened for either stx1 or stx 2 gene AND –eae gene
  - Further screened for wzx serogroup
  - If positive for stx1 or stx2, eae gene and wzx serogroup reported as potential positive

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

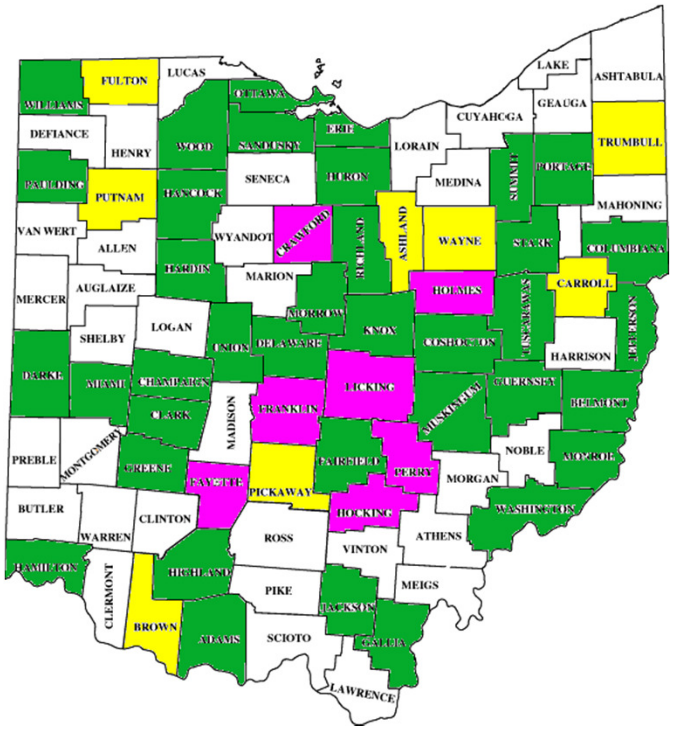
Methodology (continued)

- Any potential positives went through a cultural confirmation process
  - Immunomagnetic separation (IMS) beads
  - Plated on differential agars
    - 10-20 typical colonies selected
  - Rescreen on BioGX or BAX panels
  - Screen using antisera (agglutinate)
  - Biochemical identification via VITEK
- If positive through each step: reported as positive

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Study Population

- Fully-inspected beef slaughter facilities in Ohio
  - 76 beef slaughter facilities in Ohio
  - 69 facilities participated in study
  - 50,500 beef slaughtered annually statewide



INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Results

Summary table of Potential Positive Findings by Method

METHOD	"BIG 6" SEROTYPES					
	O145	O111	O26	O45*	O121*	O103
BioGX			2	1	1	
BAX	1		2		3	

* NO IMS BEADS AVAILABLE

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Results (continued)

- Limitations
  - Novobiocin issues (sample prep)
  - Issues with software (both)
  - Background interference (BioGX)
  - No eae gene identification (BioGX)
  - Lack of specificity (BAX)
- Difficulty in identifying 'typical' colonies for confirmation

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Conclusions

- Due to test kit weaknesses, some questions remain unanswered.
- Regulators are not ready to implement mandatory testing.
- Too many shortcomings with screening and confirmation methods.
- More validation and real-life analyses are needed to identify and address weaknesses.

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Recommendations

- More time to research and validate.
- FSIS should work with or share materials with state labs.
- Include other known serotypes of pathogenic E. coli
  - O104
- Retest Ohio beef slaughter operations

INTERNATIONAL FOOD PROTECTION TRAINING INSTITUTE

Acknowledgments

- Dr. Joanne Brown, Mentor
- International Food Protection Training Institute (IFPTI)
- Maya Achen and Tina Reed, ODA CPL
- ODA Division of Meat Inspection
- IFPTI 2011 Fellows

References

- Center for Disease Control. (2012). *Outbreaks by pathogen: Escherichia coli (E. coli)*. Retrieved from: http://www.cdc.gov/outbreaknet/outbreaks.html#pathogen
- Eblen, D. (2007). USDA, FSIS, OPHS. *Public health importance of non-O157 Shiga toxin-producing Escherichia coli (non-O157 STEC) in the US food supply*. Retrieved from: http://www.fsis.usda.gov/PDF/STEC_101207.pdf
- Scharff, R. (2010). Health related costs from foodborne illness in the United States. Produce Safety Project. Retrieved from: http://www.producesafetyproject.org/admin/assets/files/Health-Related-Foodborne-Illness-Costs-Report.pdf-1.pdf
- (2011). *MLG 5B.01 Detection and isolation of non-O157 Shiga-toxin producing Escherichia coli (STEC) from meat products*. USDA FSIS Laboratory Guidebook. Retrieved from: http://www.fsis.usda.gov/PDF/Mlg_5B_01.pdf

Questions?