SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI*

Dr. Randy Phebus

E. coli in the Food System:

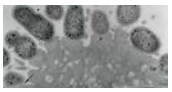
- How 7 years of the USDA Coordinated Agricultural Project (CAP) grant has improved understanding and management of the deadly STEC pathogens in the beef chain
- Sampling of STEC-focused knowledge generation across the entire food system

PURPOSE

1. Define and describe Shiga toxin-producing *Escherichia coli* (STEC).
2. Provide the scope, incidence, and impact of STECs in foodborne illness.
3. Present current research on STECs.
4. Discuss mitigation of risk for STEC, with emphasis on the role of consumers and foodservice.

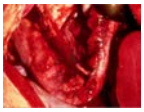
OBJECTIVES

- STEC are a type of pathogenic *E. coli* that produces a potent toxin called Shiga toxin (Stx), also known as verotoxin or verocytotoxin.
- Stx causes blood vessel damage and plays a key role in other events that result in hemorrhagic colitis (bloody diarrhea), and a type of kidney failure called hemolytic uremic syndrome (HUS) in human patients.
- Strains isolated from human patients with hemorrhagic colitis and/or HUS, and isolates positive for both *stx* and *eae* (intimin) genes are known as enterohemorrhagic *E. coli* (EHEC).
- EHEC, including *E. coli* O157:H7, are the number one cause of acute end-stage kidney failure in children.



Other key virulence determinants important in infection (especially *eae* gene), and Stx 2 is more potent than Stx 1.

Hemolytic uremic syndrome (HUS)



Hemorrhagic colitis (bloody diarrhea)

Thrombotic Thrombocytopenic Purpura (TTP)

STEC CAUSE HORRIFYING DISEASE

- Prior to family BBQ in 2007, Stephanie Smith remains in a wheelchair, fighting to walk -- and dance -- once again - She ate an *E. coli* tainted hamburger
- 2010 financial settlement with the ground beef manufacturer

NOT ONLY DIARRHEA

Mail Online

Boy, five, died hallucinating in agony after catching E-coli in school meal

By Lucia Ballard
UPDATED: 02:57 EST, 24 November 2010

A boy of five died in agony – screaming and hallucinating – after eating school dinners containing meat contaminated with E.coli, an inquest heard.

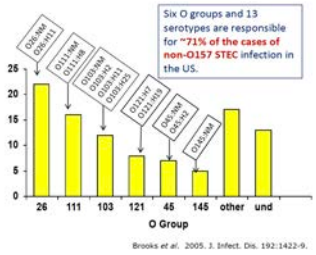
Mason Jones ate gammon and turkey supplied by a local butcher only two weeks after switching from packed lunches.

Yesterday his devastated mother told how the meals had 'utterly destroyed' her son, who died in hospital during Britain's second largest E.coli outbreak.

- One of 158 school children and adults in Britain's 2nd worst *E. coli* O157:H7 outbreak in 2005
- 31 hospitalized, Mason died.
- Butcher William Tudor jailed 1 year for breaching food processing hygiene laws
- Supplied contaminated meat to 44 schools

"E. coli is not just a tummy bug, with sickness and diarrhoea. It completely kills organs, it utterly destroyed Mason's insides." Sharon Mills (mom)

DEATH



1994 USDA declared *E. coli* O157H7 adulterant in raw non-intact raw beef products.

2011USDA declared 6 other serogroups to beef adulterant list

"Big 6"
"Big 7"

STEC O26, O45, O103, O111, O121, O145 ARE ADULTERANTS IN RAW, NON-INTACT BEEF

STEC	O157	Non-O157	Total
All illnesses	96,000	169,000	265,000
Food borne	63,000	113,000	176,000
Hospitalizations	3,300	400	3,700
Deaths	30	0	30

Food borne illnesses account for about 66%

INCIDENCE OF STEC ILLNESSES IN THE US (2000-2008)

Increased Recognition of Non-O157 Shiga Toxin-Producing *Escherichia coli* infections in the United States:
Epidemiologic Features and Comparison with *E. coli* O157 Infections

Incidence of Non-O157 STEC infections	
2000	0.12 per 100,000 population
2010	0.95 per 100,000 population

O26 (26%), O103 (22%), O111 (19%), O121 (6%), O45 (5%), and O145 (4%)

FOODBORNE PATHOGENS AND DISEASE
Volume 10, Number 5, 2013
© Mary Ann Liebert, Inc.
DOI: 10.1089/fpd.2012.1401

INCIDENCE OF STEC ILLNESSES IN THE US (2000-2010)

Most Outbreaks of EHEC O157 Infection are Foodborne

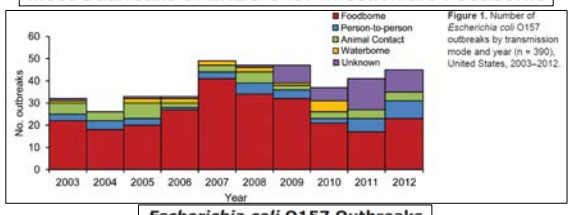
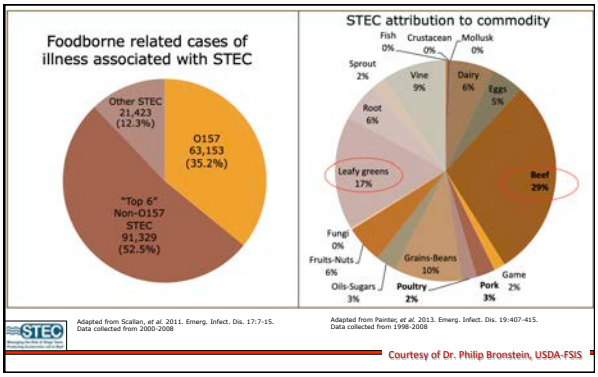


Figure 1. Number of *Escherichia coli* O157 outbreaks by transmission mode and year (n = 390), United States, 2003–2012.

***Escherichia coli* O157 Outbreaks in the United States, 2003–2012**

Katherine E. Heiman, Rajal K. Mody, Shacara D. Johnson, Patricia M. Griffin, L. Hannah Gould
Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 21, No. 8, August 2015 pp. 1293-1301



Adapted from Scallan, *et al.* 2011. Emerg. Infect. Dis. 17:7-15. Data collected from 2000-2008

Adapted from Painter, *et al.* 2013. Emerg. Infect. Dis. 19:407-415. Data collected from 1998-2008

Courtesy of Dr. Philip Bronstein, USDA-FSIS

"Shiga-toxigenic *Escherichia coli* (STEC) in the Beef Chain: Assessing and Mitigating the Risk by Translational Science, Education and Outreach"

United States Department of Agriculture
National Institute of Food and Agriculture
Agriculture and Food Research Initiative
Award Number 2012-68003-30155

THE BEEF CHAIN

$25 Million over 7 year period (awarded 1/1/2012)

Objective Areas:

1. Detection
2. Ecology/Biology
3. Interventions
4. Risk Assessment
5. Education/Outreach

Long Term Goal:

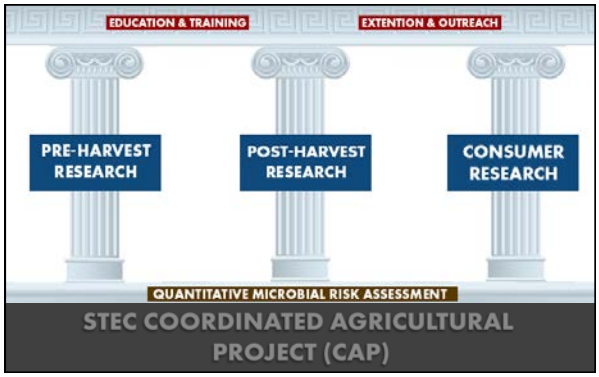
Reduce occurrence and public health risks from STEC-8 (serotypes O26, O111, O103, O121, O45, O145, O157:H7 and O104:H4) in beef using a quantitative microbial risk assessment platform

Gate to Plate

Farm to Fork

Conception to Consumption

USDA NIFA AFRI COORDINATED AGRICULTURAL PROJECT



STEC COORDINATED AGRICULTURAL PROJECT (CAP)

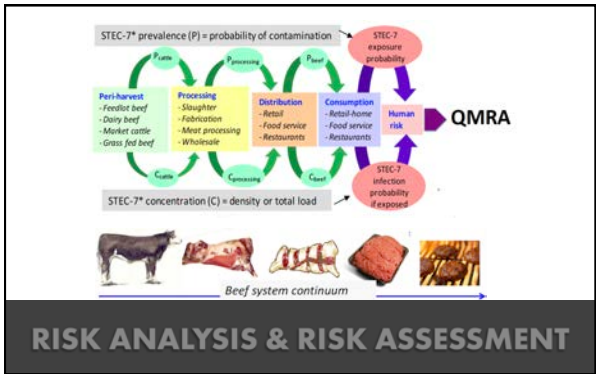
Pillar 1 - Live cattle & beef producers

Pillar 2 – Slaughter, fabrication, processing & processors

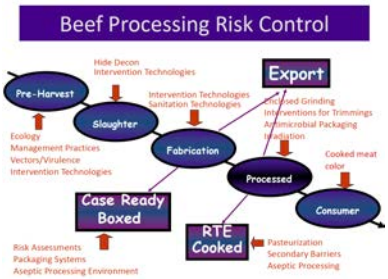
Pillar 3 - Retail, food service & consumers

Objective 1 – Detection: reagents, sampling, assays, technology, partnerships
Objective 2 – Biology: microbiology, ecology, epidemiology, modifiable risk, best targets
Objective 3 – Interventions: STEC lethality value, feasibility, cost-benefit, impacts
Objective 4 – Risk analysis: risk assessment (QMRA)
Objective 5 – Education, outreach, and evaluation: beef chain and general food safety

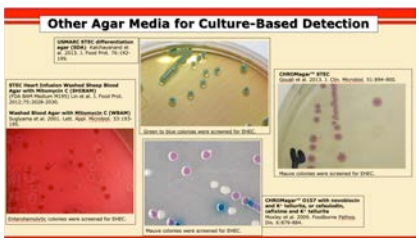
STEC CAP OBJECTIVES



RISK ANALYSIS & RISK ASSESSMENT

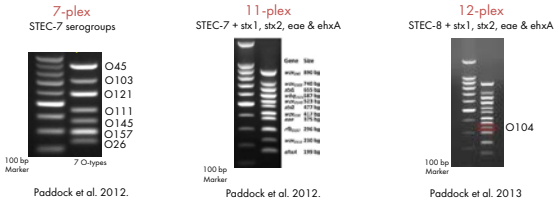


BEEF PROCESSING RISK CONTROL



Selective Enrichments
Chromogenic Agars
Spiral Plating Method
Genetic-based Testing
Screening vs Confirm
Feces vs Meat/Food
Injury Recovery

OBJECTIVE 1

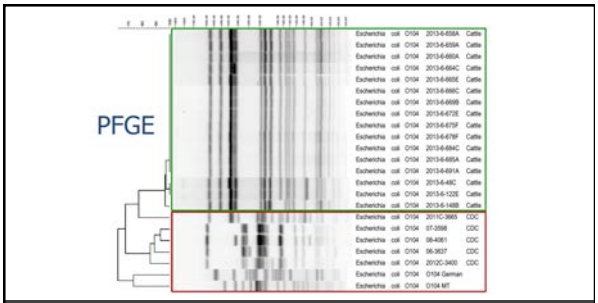


OBJECTIVE 1

Genes	No. of samples (n=757)	
	Before enrichment, %	After enrichment, %
wzx$_{O104}$ (O104-antigen flippase) ♦	5.0%	46.1%
terD (Tellurite resistance) ♦	44.8%	82.3%
stx$_1$ (Shiga toxin 1)	3.8%	27.4%
stx$_2$ (Shiga toxin 2) ♦	20.6%	66.8%
ehxA (Enterohemolysin)	48.8%	96.3%
eae (Intimin)	14.8%	72.5%
fliC$_{H4}$ (H4-specific flagellum) ♦	26.4%	86.9%
aggA (pilin subunit of aggregative adherence fimbria 1) ♦	0	0

♦Present in the German outbreak strain

PREVALENCE OF E. COLI O104: 8-GENE PCR VS. CULTURE

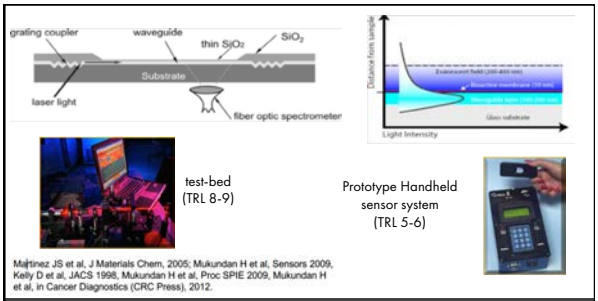


CDC – HUMAN O104:H7 STRAINS FROM CDC

US cattle O104 strains are different from the German outbreak strain

- Multiplex PCR did not detect stx 2 nor AggA
- Phylogeny by whole genome microarray did not group together with the German strain
- PFGE types are very different from the German strain, or other human strains

PREVALENCE OF E. COLI O104: 8-GENE PCR VS. CULTURE



Martinez JS et al, J Materials Chem, 2005; Mukundan H et al, Sensors 2009, Kelly D et al, JACS 1998, Mukundan H et al, Proc SPIE 2009, Mukundan H et al, in Cancer Diagnostics (CRC Press), 2012.

A WAVEGUIDE-BASED BIOSENSOR

Epidemiology of STEC in Cattle Production Systems

OBJECTIVE 2

Meta-Analysis Conclusions

- Limited non-O157 serogroup and virulence gene data in cattle
- North America yielded the highest serogroup, STEC, and EHEC estimates
- Worldwide [serogroups O26 and O103 were the most frequently detected]

Identified data gaps in published literature:

1. Data needed for prevalence of non-O157 serogroups and their virulence genes in peri-harvest cattle feces, hides, and carcasses in different cattle types (fed beef, fed dairy, cull beef, cull dairy cattle and veal calves)
2. Concentration data urgently needed to assess human exposure risks
3. Scarce data on feco-hide-carcass microbial contamination pathway
4. Limited data on potential "drivers" of prevalence (e.g., geographical region, season, production system) or variability (e.g., region, feedlot, pen)

OBJECTIVE 2

Need to Know …

- Prevalence & genetic characteristics of *E. coli* O104 in cattle at feedlots and harvest
- Data gaps on prevalence & concentration of non-O157 STEC in literature exposed through meta-analysis
- Seasonal aspects of non-O157 STEC prevalence in feedlot cattle
- Regional, feedlot and pen-level variability in prevalence of non-O157 STEC in fed cattle
- Prevalence & concentration of STEC on hides of fed and cull cattle, and on resultant beef carcasses
- Quantification of microbial transfer from hides to carcasses during beef harvesting operations
- Prevalence of STEC in veal calves

OBJECTIVE 2

- Role of house flies in ecology of STEC-7 in confined cattle environments
- Mathematical models of transmission dynamics of STEC in cattle
- Analysis and interpretation of bacterial communities (microbiome) within hide swab and fecal samples of cattle at harvest—association of STEC with shifts in communities
- Genetic characterization of STEC O103 isolates from cattle & humans
- Gene expression of STEC O103 during bacterial interaction with bovine rectal epithelial cells
- Prevalence & characterization of *E. coli* O157:H7 and non-O157 STEC recovered from retail ground veal in Mid-Atlantic region

OBJECTIVE 2

Summer and winter prevalence of STEC O26, O45, O103, O111, O121, O145, and O157 in feces of feedlot cattle

Diana M. Dewsbury, David G. Renter, Pragathi B. Shridhar, Lance W. Noll, Xiaorong Shi, T.G. Nagaraja, and Natalia Cernicchiaro

Foodborne Pathogens and Disease, 2015

OBJECTIVE 2

- Some non-O157 serogroups relatively common in cattle feces (e.g., O103); others were not (e.g., O111)
- Few non-O157 STEC were recovered in summer months; most serogroup positive samples did not harbor virulence genes
- No STEC were isolated during the winter months
- Seasonal differences were observed
- Fecal shedding was highly variable between pens of cattle

OBJECTIVE 2: CONCLUSIONS

Regional, feedlot and pen-level variability in prevalence of non-O157 STEC

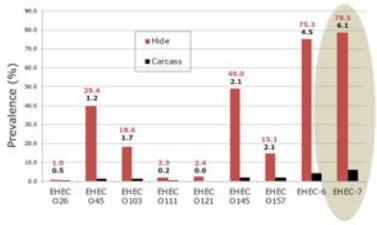
Charley Cull, David G. Renter, Diana M. Dewsbury, Pragathi B. Shridhar, Lance W. Noll, Xiaorong Shi, Samuel Ives, T.G. Nagaraja, And Natalia Cernicchiaro

OBJECTIVE 2

- Order of STEC prevalence: O157, O103, O145, O45, O26, O111 and O121
- 100% of feedlots (n=8) and 62% of pens (n=126) had feces positive for O157 STEC, with 100% of feedlots and 23% of pens positive for non-O157 STEC
- No significant differences between states; no statistically significant feedlot-level risk factors (e.g., demographic, dietary, management)

OBJECTIVE 2: CONCLUSIONS

Comparison of Hide and Carcass Prevalence of EHEC-7 in Commercial Feedlot Cattle at Harvest – Summer 2013

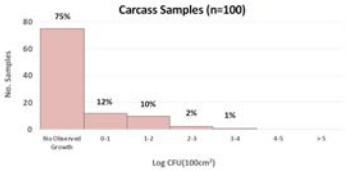


OBJECTIVE 2

Prevalence and concentration of STEC and surrogate microorganisms on hides of fed and cull cattle

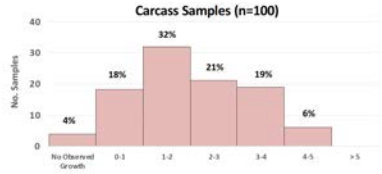
David G. Renter, Sam E. Ives, Lance W. Noll, Pragathi B. Shridhar, and T.G. Nagaraja

OBJECTIVE 2



Week	No. of sample positive, Log CFU/100 cm²						
	No Observed Growth	0-1	1-2	2-3	3-4	4-5	>5
1	15	3	4	2	1	0	0
2	25	0	0	0	0	0	0
4	21	4	0	0	0	0	0
5	14	5	6	0	0	0	0
Total	75	12	10	2	1	0	0

ENUMERATION OF COLIFORMS FROM PRE-EVISCERATION



Week	No. of sample positive, Log CFU/100 cm²						
	No Observed Growth	0-1	1-2	2-3	3-4	4-5	>5
1	0	0	10	2	8	5	0
2	1	6	6	7	4	1	0
4	2	5	5	8	5	0	0
5	1	7	11	4	2	0	0
Total	4	18	32	21	19	6	0

ENUMERATION OF COLIFORMS FROM POST-EVISCERATION

- Biosafety Level 3-Ag Research
- Suites for Plant, Animal, Food, Molecular Biology, Insectary
- One of a kind research facility in the world!

- ✓ Biology of pathogens
- ✓ Diagnostic technology development
- ✓ Food safety & defense during Processing
- ✓ Contamination control and mitigation
- ✓ Real pathogens & surrogates

OBJECTIVE 3

Difference Surfaces – Different Stresses – Different Results

Biosecurity Research Institute (BRI)

OBJECTIVE 3

Electrostatic Carcass Spray Cabinet

Chad Carcass Spray Cabinet
180°F Water/Chemical

Carcass Spray Chill w/ Chemical Appl.

Supporting Attached Microbiology Lab

Fully Equipped Meat Fabrication/Processing Floor

BSL-3 SLAUGHTER & MEAT PROCESSING SUITE AT BRI

- Effect of non-digestible fiber and wet distillers grains on feedlot cattle shedding of STEC (UNL experimental feed yard)
- Efficacy of different approved antimicrobial interventions (e.g. organic acid sprays) in reducing STEC on beef carcasses, fabricated cuts, and head meats (sequencing and multi-hurdle applications)
- Electrostatic spray technology to improve bacterial kill and save water/energy
- Validation of fermentation & heating of dry-fermented sausages to control STEC
- Thermal inactivation of STEC within cubed beef steaks and veal cordon bleu

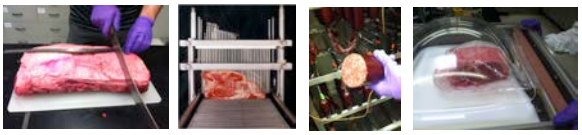
200 ml fluorescent dye applied by ESS to carcass

OBJECTIVE 3

- Sprayed Lethality in Container (SLIC) method to deliver antimicrobials for STEC onto vacuum packaged beef subprimals
- Translocation & thermal inactivation of STEC in blade tenderized or vacuum tumbled raw beef cuts (e.g. roasts, prime rib)
- Effect of high pressure processing on survival of STEC in beef meatballs and summer sausage
- Effect of deep frying and conventional oven cooking methods on inactivation of STEC in meatballs (veal and/or beef-pork-veal)

OBJECTIVE 3

BEEF PROCESSING INDUSTRY / TECHNOLOGY INDUSTRY / ACADEMIA / GOVERNMENT
MASSIVE SCALE COLLABORATIVE EFFORT



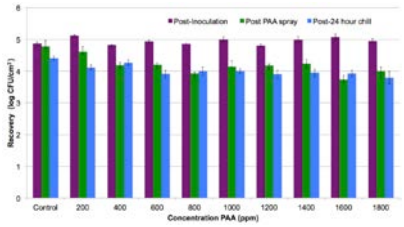
OBJECTIVE 3

STEC CAP VEAL INIATIVE

- Hide-on carcass traditional and novel intervention validations (Fresno State)
- Dressed carcass chemical spray intervention validations (K-State)
- Breaded veal cutlets cooking and cordon bleu STEC risk profiling studies
- STEC prevalence and characterization in retail ground veal (4 states)
- K-State Beef Cattle Institute veal producer and processor training modules

OBJECTIVE 3

Peracetic Acid Concentration Effects on STEC Reductions in Vacuum Packaged Beef Subprimals



OBJECTIVE 3

Comparing STEC Survival in Low- (7%) and High-Fat (30%) Ground Beef During Heating

Findings:

- No differences in survivability across "Big 8" STEC serogroups
- Slightly greater survival of STEC in high-fat ground beef
- Cooking times and temperatures deemed effective for inactivating *E. coli* O157:H7 equally effective for the other 7 serogroups

OBJECTIVE 3

Refining Food and Nutrition Science Education through Piloting and Capstone Development

K-State University
University of Nebraska
USDA ERRC

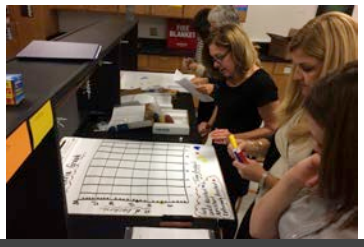
OBJECTIVE 5

Project Based Learning

- Use the scientific method to develop and conduct research
- Engage high school students in food science, nutrition science and food safety research
- Encourage students to pursue food, nutrition science careers

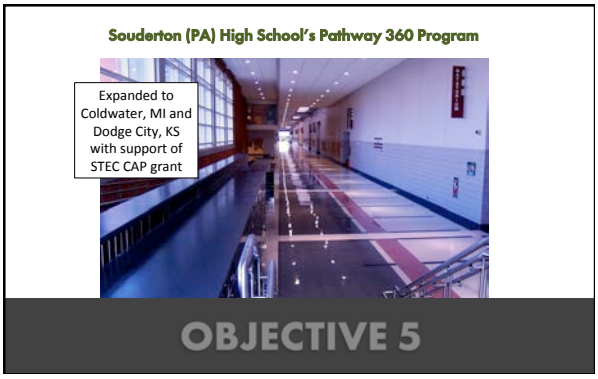
OBJECTIVE 5

Exponential Growth Lab



OBJECTIVE 5


KANSAS STATE UNIVERSITY
http://www.k-state.edu/fns/
Food and Nutritional Sciences
Food and Nutrition Science Curriculum
A USDA-NIFA project
Food and Nutrition Science Curriculum
Home
Course 1: Food Production, Science, and Health
Course 2: Food Science
Course 3: Nutrition Science
Course 4: Food and Nutrition Science Research and Development
Middle School Curriculum
Megaburgerz Comic Series
Communications and Agricultural Education
The U.S. Department of Agriculture-National Institute of Food and Agriculture (USDA-NIFA) Coordinated Agricultural Project (CAP) grant, titled Shiga-toxigenic Escherichia coli (STEC) in the Beef Chain: Assessing and Mitigating the Risk by Translational Science, Education and Outreach, seeks to significantly advance evidence- and action-based beef food safety knowledge to protect public health. This project is targeting the eight most important STEC serotypes that cause human illness in the U.S.
The grant has 5 objectives; Objective 5 concerns Risk Management and Education and focuses on training the current and future workforce and consumers to control STEC. To train the current and future workforce, the team is developing curriculum for secondary education programs at the high school level to gain access to the students who are in these classrooms.
The multi-year curriculum is comprised of four courses, including a Capstone course, and has been developed for middle and high school students. The curriculum has an inquiry-based approach with integrated instructional aids for teachers and their students. Each course consists of four units which each have daily lessons plans.


Souderton (PA) High School's Pathway 360 Program
Expanded to Coldwater, MI and Dodge City, KS with support of STEC CAP grant
OBJECTIVE 5


Souderton Increase in STEM Enrollment • 159% since 2010
OBJECTIVE 5


Assessing handling practices and perceptions of mechanically tenderized beef
NC STATE UNIVERSITY
NC State University A&T State University COOPERATIVE EXTENSION
Empowering People - Providing Solutions
Renee R. Boyer, Benjamin Chapman, Lily Yang, Nicole Arnold, Minh Doung
OBJECTIVE 5

Labeling requirements – May 2016

– To include descriptive designation
– Easy to read
– Must include validated cooking instructions

- Cooking method
- Minimum internal temperature
- Hold times
- Temperature must be measured

LABELING RULES

NC State Current Project:

- ✓ Collect data related to consumer knowledge & practices of MTB products
- ✓ Make recommendations for how to best communicate risks associated with mechanically tenderized beef to consumers – Intervention Methods
- ✓ Collect data for STEC- beef risk assessment

ULTIMATE GOAL OF OUR WORK

Comparing Methods of Delivery of Food Safety Information to Consumers

- Positive Deviance (PD) focus group method = novel educational intervention that allows participants to discuss their food handling behaviors and decide to try recommended practices modeled after people like themselves.
- Compared PD to personal storytelling and reading standard materials with 89 pregnant women and 93 diabetics.
- Assessed self-reported food safety knowledge, behavioral changes, and hygiene practices pre- and post-intervention through survey
- Found that PD had higher knowledge scores and adopted more safe handling recommendations. Suggests that food safety education is most effective when delivered in a supportive discussion format.

Dr. Christine Bruhn and Yaohua Feng … UC-Davis

OBJECTIVE 5

TV Celebrity Chefs as Role Models for Consumers' Safe Food Handling in the Home

- TV chefs frequently fail to follow recommended food-handling behaviors.
- Study investigated food-handling practices of 4 celebrity chefs (59 shows scoring cook, clean, chill and separate), and consumers' and culinary students' attitudes toward mishandling.
- Culinary students believed that chefs should serve as positive role models.
- Consumers viewed celebrity chefs as role models, utilized information transmitted during cooking shows, and practiced behaviors they observed.
- Celebrity chefs' poor food-handling practices could increase risk of foodborne illness associated with food prepared in the home.

Dr. Christine Bruhn and Yaohua Feng … UC-Davis

OBJECTIVE 5

Assessment of Risk Communication about Undercooked Hamburgers by Restaurant Servers (Secret Shopper Study)

- It is the duty of food establishments to disclose and remind consumers of risk when ordering undercooked food such ground beef (FDA Food Code 2013.
- Explored risk communication behaviors of food establishment servers using secret shoppers to visit 265 restaurants in 7 states and ordering medium rare burgers.
- Majority of servers reported an unreliable method of doneness (77%), and 66% of servers provided incorrect (according to Food Code) food safety information to consumer.
- Results demonstrate major gaps in server risk knowledge and support more effective food safety training if servers are to be risk communicators and lead to informed decisions by consumers.

Dr. Ben Chapman and Ellen Thomas… NC State

OBJECTIVE 5

Consumer Food Handling Practices Lead to Cross-contamination

Purpose (Sneed et al., 2015): Determine impact of "Food Safety Families" clean and separate messages on cross-contamination behaviors of consumers in the kitchen.

- 123 participants randomly assigned to a control group, or one of two food safety message groups
- All three groups videoed preparing home meal from raw poultry or ground beef, coupled with a hand-cut fruit salad
- Monitored contamination spread during meal prep microbiologically (*Lactobacillus casei*) and scored behaviors (video)
- 90% of salads were contaminated and 24% were highly contaminated (levels slightly lower for food safety messages groups)
- Handwashing scores lower for control group
- Towels were frequently handled by participants and were a primary source of contamination spread
- External food safety cues had a slight positive effect on behaviors
- Regardless of group, most participants used procedures resulting in kitchen/food cross-contamination

Phebus/Industry Flour Safety and Bakery Products Safety Initiative

- *Salmonella* and STEC are potential risks in raw flour (including cake mixes) and has led to outbreaks and recalls
- Flour easily contaminates the kitchen (home, food service, processing plant) environment and can re-contaminate baked goods.
- Working with industry millers to engineer a method of decontaminating wheat prior to milling
- Working with the baking industry to assess various thermal manufacturing protocols for diverse products for control of *Salmonella* and STEC
  - ✓ Baking and frying as an effective kill step (5-log cycle reductions)
  - ✓ D- and z-value generation for *Salmonella* and STEC during heating
  - ✓ Generating free online "Kill Step Calculators" for industry to prove/ confirm lethality of proprietary baking processes
  - ✓ Determination of survival period of *Salmonella* and STEC in dry stored flour

THANK YOU!

Randall Phebus
Professor of Food Safety
Food Science Institute
Kansas State University
phebus@k-state.edu

www.stecbeefsafety.org

https://www.asi.k-state.edu/about/people/faculty/phebus/

FoodHandler® SafeBites

JOIN US FOR FUTURE WEBINARS!

WEDNESDAYS @ 1PM (CT)

2019 WEBINARS

MAR 27	Workforce Development in Food Safety
MAY 15	Food Storage
SEPT 11	Communication
OCT 16	Controlling Costs
NOV 13	Preparing For An Emergency/Disaster

Each are preapproved for 1 hour of Continuing Education Credit by the School Nutrition Association (SNA) and the Certifying Board for Dietary Managers (CBDM)

FoodHandler® SafeBites

COMPLETION CERTIFICATES

Certificates will be mailed out within 5-7 business days, following today's webinar.

For more information and to register:

WWW.FOODHANDLER.COM/EDUCATION-TRAINING

WEBINAR RESOURCE

INFORMATION & REGISTRATION

Videos

- Handwashing
- Why To Glove
- When To Glove
- How To Glove

Documents

- Daily Temperature Logs
- Temperature Chart For Safe Food
- Refrigerator Storage Chart
- Food Safety Doesn't Happen By Accident

FOODHANDLER RESOURCES

Past Blogs

- Politics of Food Safety
- Holiday Health and Food Safety
- Employee Health
- Norovirus

Upcoming Blogs

- Employee Health & Reportable Illness
- Active Managerial Control*
- Coaching & Training Staff
- Productivity

FOODHANDLER RESOURCES

Please Send Us Your Questions Or Comments At:

FOODSAFETY@FOODHANDLER.COM

HOW DID WE DO?

FEEDBACK AND COMMENTS