2011 Beef Industry Safety Summit

Research Update

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Introduction

"We used to see, touch and smell our way to safe food," said Dr. Elizabeth Hagen during her keynote address. "Research and technology have helped us catch up and make significant progress in recent times. Science is our anchor."

That sentiment rings true for the beef industry as scientific solutions are the cornerstone of efforts to improve beef safety. The 2011 Beef Industry Safety Summit included an update on some of the latest advancements in safety research from leading researchers in the field.

THE BEEF INDUSTRY SAFETY SUMMIT—ONE OF THE MOST RESPECTED SAFETY MEETINGS IN THE INDUSTRY— IS COORDINATED BY THE NATIONAL CATTLEMEN'S BEEF ASSOCIATION (NCBA) ON BEHALF OF THE BEEF INDUSTRY FOOD SAFETY COUNCIL (BIFSCO) AND IS PARTIALLY FUNDED BY THE BEEF CHECKOFF.

The Effect of Sample Size and Matrix on the Ability of Rapid Methods to Detect E. coli *O157:H7*

Mick Bosilevac, USDA-ARS, U.S. Meat Animal Research Center

Testing methods for detecting *E. coli* O157:H7 are constantly evolving, and it is important to validate them to ensure accuracy. Researchers evaluated how changes in sample size, volume of test media and the duration of time that bacteria were allowed to develop in enrichment formulas affected the detection of *E. coli* O157:H7.

The results showed increasing sample size or decreasing the volume of test media affected the accuracy of detecting *E. coli* O157:H7 at 8, 12, and 16 hours of incubation. Bacterial detection at 12 hours and 16 hours could be improved through the use of more selective media. Liver samples required a phosphate-buffered medium for the proper detection of *E. coli* O157:H7. The results of this research can provide guidance on the use of modified enrichment media, and the length of time needed to detect *E. coli* O157:H7 by different tests in different sizes and types of samples, and can also be used as supporting documents for in-plant testing programs.

Understanding How Salmonella *Potentially Evades Beef-Carcass-Processing Interventions*

Dayna M. Brichta-Harhay, USDA-ARS, U.S. Meat Animal Research Center

While significant strides have been made by the beef industry to address *Salmonella* prevalence on beef carcasses at harvest, low-level contamination continues to be of concern. *Salmonella's* ability to adapt to safety interventions, and the internalization of bacteria within lymph tissue, are two possible contributors to the low-level persistence.

Researchers evaluated the prevalence of *Salmonella* in subiliac lymph nodes obtained from cattle at harvest and also characterized the gene expression response of multi-drug-resistant (MDR) *Salmonella* Newport to various carcass-processing interventions. *Salmonella* was present in 8.7 percent of the lymph nodes analyzed. Since *Salmonella* was readily recovered from these tissues, the researchers concluded fat trim may be a point source for *Salmonella* entry into ground beef products. Further research should be conducted to eliminate the presence of *Salmonella* in the lymph nodes of healthy cattle.

To help explain the molecular strategies that *Salmonella* employ to survive multiple-hurdle, beef-carcass-processing interventions, genes were evaluated. Genes demonstrating the greatest change in expression included those with roles in heat shock response, DNA repair and a number of regulatory systems that govern the *Salmonella* virulence response. These genetic adaptations may be exploited to reduce the presence of *Salmonella* in beef products.

Dietary Orange Peel and Pulp Can Reduce E. coli *O157:H7 Populations in the Intestinal Tract of Ruminants*

Nathan Krueger, USDA-ARS, Food and Feed Safety Research Unit

Orange peel and pulp is available at low prices in citrus-producing regions and is often used as a low-cost feedstuff for beef and dairy cattle. Orange peel and pulp and other citrus fruits contain essential oils such as limonene and linalool that are toxic to bacteria and exhibit an antioxidant effect in host animals. Previous research demonstrated orange peel and pulp reduced *E. coli* O157:H7 and *Salmonella* in laboratory settings. An *in vivo* study was performed using sheep as a test model to determine the effect of orange peel and pulp as either 0, 5 or 10 percent of the total ration on the reduction of *E. coli* O157:H7. Fecal shedding was reduced, as well as the population of *E. coli* O157:H7 throughout the gastrointestinal tract of the test animals. The most notable reduction was in the rumen at both the 5 percent and 10 percent diet concentrations.

Corn-Based Distillers' Grain and the Burden of E. coli *O157:H7 in Commercial Feedlot Settings and the Development of a Semi-Quantitative Method to Estimate Concentration of* E. coli *O157:H7*

Evan Chaney, Texas Tech University

Researchers quantified the potential relationship between the use of distiller's grains (DG) and *E. coli* O157:H7 pathogen loads in actual feedlot settings, compared two different bacterial sampling methods, and assessed how samples are impacted by duration of storage. Two experimental groups of cattle were fed either a 15 percent DG or 8 percent DG ration. The results indicate a higher prevalence of *E. coli* O157:H7 associated with diets including a higher concentration of DG. The semi-quantitative sampling method proved to be a potential alternative that is less expensive and time consuming; however, fecal samples should be analyzed within two weeks of collection to ensure optimal results.

The Effect of Feeding Pasteurized or Non-Pasteurized Waste Milk on Fecal Populations and Prevalence of Salmonella *in Dairy Calves*

Russell Farrow, USDA-ARS, Food and Feed Safety Research Unit

Dairy producers often use waste milk (commonly from cows undergoing veterinary treatment) as a feed source for young calves as it can't be sold commercially. In order to reduce the potential transmission of pathogens to the calves, some producers pasteurize the waste milk prior to feeding. Researchers evaluated the effect of pasteurization on *Salmonella* in dairy calves and found the practice did not have a significant impact on reducing *Salmonella* over the course of the study. While results of this study did not find any significant effect of waste-milk pasteurization on *Salmonella* prevalence in dairy calves, researchers do not want to discourage the practice as it may have other beneficial effects on important calf pathogens such as *Mycobacterium paratuberculosis* that can be present in milk.

A Unique Genotype Conferring Resistance to Salmonella *in Certain Red-and-White Holstein Steers*

Steve Carlson, Iowa State University

Anecdotal evidence was explored to determine if certain lines of Red-and-White Holstein cattle bear a unique genotype conferring resistance to *Salmonella*. Single nucleotide polymorphism (SNP) analysis revealed one consistent gene mutation that inactivates the expression of host cell protein exploited by *Salmonella* during the infection process. Further, an *in vitro* study revealed *Salmonella* were not able to survive in white blood cells obtained from Red-and-White Holstein steers that were homozygous for the SNP of interest. The effect was dramatically less pronounced in all heterozygous animals, homozygous female Red-and-White Holsteins, and homozygous Black-and-White Holsteins of either gender. *In vivo* studies supported these results. Eighteen days post-infection, clinical manifestations of salmonellosis were not observed in Red-and-White Holstein steers homozygous for the SNP. *Salmonella's* ability to colonize was poor in the homozygous group compared to the other gene combinations. This research shows certain lines of cattle have the genetic potential to be resistant to *Salmonella* infection.

Antibiotic and Disinfectant Susceptibility Profiles of E. coli *O157:H7 From Cattle Feces, Hide, Carcass, and Ground Meat Isolates From the United States*

Ross Beier, USDA-ARS, Southern Plains Agricultural Research Center

Researchers evaluated the disinfectant and antibiotic susceptibility profiles of 344 *E. coli* O157:H7 isolates from cattle feces, hide, carcass and ground meat from different parts of the United States. All of the *E. coli* O157:H7 isolates were susceptible to fluroquinolones. A low incidence of antibiotic resistance was observed in 16 antibiotics commonly tested as part of the National Antimicrobial Resistance Monitoring System (NARMS). High multi-drug resistance was observed in two cull cow isolates and in two ground meat isolates, all four to the same eight antibiotics. Susceptibility of the *E. coli* O157:H7 isolates was also measured against 21 disinfectants and disinfectant components. All of the isolates were susceptible to troclosan, but 69 were resistant to chlorhexidine, and benzalkonium chloride. Acetic, citric and lactic acids were effective in that order of potency. Of the 24 bacteria that demonstrated chlorohexidine resistance, nine of the bacteria were also resistant to antibiotics. A high incidence (6.1%) of resistance was observed in the bacteria to quaternary ammonium chlorides, which are used as surface disinfectants and sanitizers in dairies, restaurants and food processing plants. This could be problematic unless properly addressed, as it could allow some bacteria to escape disinfection.

Sensitivity of Mycobacterium bovis *to Common Beef Processing Interventions*

Mick Bosilevac, USDA-ARS, U.S. Meat Animal Research Center

Mycobacterium bovis is the causative agent of bovine tuberculosis, which can be spread to humans through inhalation or by ingestion. *M. bovis* multiplies slowly, so infected animals may be sent to slaughter during the early stages of the disease before diagnosis. Since cattle actively shedding *M. bovis* may contaminate the beef-processing environment, the objective of this study was to evaluate multiple processing interventions and determine which may be the best means to control *M. bovis* if it is present.

Treatments with chlorine, phosphoric acid, hydrochloric acid and two different concentrations of lactic acid did not reduce the number of *M. bovis* at ambient temperatures. However, treatment with either two different concentrations of lactic acid at 50°C, peroxyacetic acid or bromine did reduce *M. bovis* by 1.5 to 2 log colony-forming units (CFU) when compared to the control. Additionally, acidified sodium chloride and hot water (≥ 75°C) reduced *M. bovis* by at least 3 log CFU.

Reduction of E. coli O157:H7 in Mechanically Tenderized Beef Strip Steaks Using Lactic Acid and Cooking

Jennifer Martin, Texas Tech University

Mechanical tenderization improves beef palatability, but may also pose an increased risk of translocating pathogenic bacteria from the surface to the interior of beef cuts. This study investigated the effect of a common fresh beef intervention, lactic acid spray (5%), on the survivability of *E. coli* O157:H7 in mechanically tenderized beef steaks managed under simulated industry conditions and cooked to various degrees of doneness.

Treatment with five percent lactic acid reduced the initial levels of *E. coli* O157:H7 on the first day of the experiment on both the lean and fat surfaces of beef strip loins. After 21 days of storage in vacuum packages, further testing revealed lactic acid didn't reduce *E. coli* O157:H7 on lean surfaces of strip loins inoculated with a high level of *E. coli* O157:H7 (10^5 log CFU/cm^2), but a reduction was noted on the fat surfaces. Lactic acid spray also reduced *E. coli* O157:H7 on the surface of steaks from needle tenderized strip loins inoculated with high levels of bacteria, but *E. coli* O157:H7 was still detected in core samples from needle tenderized steaks that were inoculated with high levels of bacteria on the surface and cooked to 55, 60 or 70°C (131, 140 or 158°F). *E. coli* O157:H7 was not detected in core samples obtained from steaks inoculated with low levels of bacteria (10^3 log CFU/cm^2), regardless of cooking temperature, thus demonstrating lactic acid's effectiveness.

Validation of Lactic Acid for Use as a Subprimal Antimicrobial Intervention

Curtis Pittman, Colorado State University

Lactic acid has been shown to reduce bacterial counts on beef carcass surfaces when used as a food safety intervention, but effectiveness when applied to the surface of chilled beef subprimals is not well documented. The objective of this study was to validate initial use of lactic acid as a subprimal intervention during beef fabrication, followed by a secondary application to vacuum-packaged product following the removal of packaging. Beef knuckles and briskets were inoculated with one of four strains of *E. coli*, and one strain of *Salmonella* and were subjected to lactic acid treatments in 16 different combinations of two different temperatures, two different pressures, two application rates and two concentrations.

Initial application of lactic acid at 22°C, pressure of 1.03 bar, 0.22 lpm and at 2.5 percent concentration resulted in the greatest total inoculated count reductions. For *E. coli* O157:H7 specifically, the greatest reduction occurred when lactic acid was applied at 22°C, 4.83 bar, 0.22 lpm and at 2.5 percent concentration.

Both the initial and secondary lactic acid treatments successfully resulted in reductions of pathogenic and non-pathogenic surrogate strains of *E. coli*, as well as natural microflora. Information from this research should be helpful to beef processors as they validate their HACCP systems.

Modeling Pre-Harvest and Harvest Interventions for E. coli O157 Contamination on Beef Cattle Carcasses

Megan Jacob, Kansas State University

Risk assessment modeling may be an effective way to evaluate the effects of multiple concurrent pre-harvest interventions for *E. coli* O157. Researchers in this study constructed a risk-assessment model with data obtained from a systematic review of scientific literature to examine the impacts of different combinations of pre-harvest and post-harvest interventions for *E. coli* O157. The researchers estimated the risk of carcass contamination conditional on pre-harvest fecal-prevalence estimates, inclusion of feed additive(s) in diet, vaccination for *E. coli* O157, transport and lairage effects, hide intervention(s) and carcass intervention(s).

The results indicated combinations of pre-harvest interventions may be particularly important for supplementing harvest interventions during periods of higher variability in fecal shedding prevalence, such as during the peak summer months. The matrix also identified future research needs, including further assessments of the relationships between fecal prevalence and concentration, as well as hide contamination and subsequent carcass contamination. Researchers are currently working to expand the model to include concentrations of *E. coli* O157, but more data relating fecal prevalence and shedding rates to hide concentration, and to carcass contamination risk and concentration, are needed to robustly estimate risk and intervention efficacy.

Consumer Adherence to Safe Handling Guidelines While Preparing Burgers and a Salad

Christine Bruhn, University of California, Davis

The objective of this study was to evaluate consumer adherence to safe handling guidelines as outlined in the government FightBAC! recommendations and the 2005 Food and Drug Administration (FDA) Food Code. Researchers filmed consumers preparing hamburgers and a salad and then evaluated the footage. Attitudes and knowledge about food safety were also assessed through a questionnaire administered after filming.

Visual appearance was the most commonly reported method for determining doneness. Checking for brown interior was the most common method, followed by juice clarity or color, and patty size and shrinkage. Seventy percent of the consumers surveyed, cooked their ground beef to the recommended internal temperature of 160°F or above, however 22 percent determined the burger was done when the temperature was below 155°F. Although 53 percent of volunteers indicated they owned a food thermometer and 33 percent said they know how to use one, only four percent actually used a thermometer. Sixty-five percent of the volunteers reported not knowing the recommended temperature for cooking ground beef, and of those that thought they knew, only 12 percent responded correctly. Seventy-six percent of consumers surveyed said they would not use a thermometer to check the doneness of burgers, believing it was not necessary. Fewer than half of the volunteers washed their hands prior to beginning food preparation and only 41 percent of handwashing events involved soap. Potential cross-contamination was common, with an average of 34 potential transfers from one surface to another as a result of contact with a potential source of contamination.

This project indicates that consumers frequently do not follow recommended food handling practices. Should raw meat contain pathogens, the probability of inadequate cooking or cross contamination is high and foodborne illness may follow.

For more information, visit

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