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Electrostatic spray cabinet evaluation to verify uniform delivery of chemical and biological solutions to pre-chilled meat animal carcasses

Abstract
Shiga toxin-producing Escherichia coli (STEC) are a group of bacteria that cause an estimated 265,000 illnesses, 3,600 hospitalizations, and 30 deaths annually in the United States. STEC are frequently associated with raw or undercooked meat products, prompting the beef industry to develop and apply various antimicrobial intervention technologies during processing operations. The application of chemical antimicrobials to carcasses and fabricated cuts using an electrostatic spray (ESS) system (Figure 1) offers several potential advantages for controlling disease-causing pathogens, including enhanced chemical deposition (coverage) profiles, reduced overspray wastage of foodgrade antimicrobials, and reduced water requirements. The objectives of this study were to (1) calibrate an ESS carcass cabinet installed at the Kansas State University Biosecurity Research Institute, (2) test the chemical deposition profile of the ESS cabinet onto a meat carcass using fluorescent dye, and (3) determine if the ESS could be used to uniformly apply a biological inoculum to a carcass to support pathogen-inoculated validation studies of different chemical intervention technologies to support the needs of the beef processing industry.

Keywords
Cattlemen's Day, 2014; Kansas Agricultural Experiment Station contribution; no. 14-262-S; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 1101; Beef Cattle Research, 2014 is known as Cattlemen's Day, 2014; Beef; Electrostatic spray; Shiga toxin-producing E. coli

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Electrostatic Spray Cabinet Evaluation to Verify Uniform Delivery of Chemical and Biological Solutions to Pre-Chilled Meat Animal Carcasses

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Introduction
Shiga toxin-producing *Escherichia coli* (STEC) are a group of bacteria that cause an estimated 265,000 illnesses, 3,600 hospitalizations, and 30 deaths annually in the United States. STEC are frequently associated with raw or undercooked meat products, prompting the beef industry to develop and apply various antimicrobial intervention technologies during processing operations. The application of chemical antimicrobials to carcasses and fabricated cuts using an electrostatic spray (ESS) system (Figure 1) offers several potential advantages for controlling disease-causing pathogens, including enhanced chemical deposition (coverage) profiles, reduced overspray wastage of food-grade antimicrobials, and reduced water requirements. The objectives of this study were to (1) calibrate an ESS carcass cabinet installed at the Kansas State University Biosecurity Research Institute, (2) test the chemical deposition profile of the ESS cabinet onto a meat carcass using fluorescent dye, and (3) determine if the ESS could be used to uniformly apply a biological inoculum to a carcass to support pathogen-inoculated validation studies of different chemical intervention technologies to support the needs of the beef processing industry.

Experimental Procedures
Calibration of the 8 ESS nozzles inside the cabinet was accomplished by testing and adjusting the air pressure and fluid flow at each nozzle. The air pressure was measured with an air flow meter (King Instrument Company, Garden Grove, CA). Two 500-mL graduated cylinders were used to measure the water flow rate (mL/minute) at each of the nozzles. A 30XR-A Auto Ranging Digital Multimeter with VolTect (Amprobe, Everett, WA) was used to measure the negative electrical charge of the fluid generated at each nozzle and used with the water flow rate to calculate a charge-to-mass ratio. To ensure adequate attraction of spray to the grounded carcass surface, a charge-to-mass ratio between -5 mC/kg and -12 mC/kg was needed.

A fluorescent dye carcass deposition test was conducted by spraying ~6.8 oz of a 1:100 concentrated dye solution (Risk Reactor IFWB-C0 Fluorescent Clear Blue Water Based Tracer Concentrate, Santa Ana, CA) within the sealed ESS cabinet containing a skinned pre-chilled pig carcass side as a model (Figure 1). The dye solution was applied to two separate one-quarter pig carcasses, then to half a pig carcass. A black light (American Fluorescent, Waukegan, IL) was used to observe the uniformity of dye deposition onto all carcass surfaces (exterior and internal body cavity; Figure 2).

A carcass inoculation study was conducted using the ESS cabinet. Stationary phase inoculum (6.3 quarts, ~8.8 log cfu/mL) was prepared using a 2-strain cocktail of non-pathogenic *Escherichia coli* biotype 1 (ATCC BAA-1429 and 1431), and ~6.8 oz of this inoculum was electrostatically sprayed onto two separate pig carcass sides.
After 30 minutes of microbial attachment, each carcass side was sampled at 8 different anatomic locations using surface tissue excision. Duplicate excised tissue samples were taken from the upper, middle, and lower regions of both the internal and external body cavity surfaces. Sponge samples were also taken at the top and the bottom of the half carcasses. A comparison of *E. coli* levels achieved at each anatomic carcass location during ESS spraying was determined by plating serial dilutions of samples using Eosin Methylene Blue agar, with incubation at 79°F for 24 hours.

**Results and Discussion**

Both the fluorescent dye test and the *E. coli* inoculation test showed highly uniform coverage. The fluorescent dye test appeared to cover all carcass surfaces in a uniform manner, including body cavity, split line, and hock areas (Figure 2). The inoculation test showed a uniform recovery of ~5–6 log cfu/cm² (100,000 to 1 million bacteria/cm²) across all anatomic regions, except a slightly lower inoculum level at the top hock area (Figure 3). The lower inoculum level at the top hock area could have been due to the difference in sampling technique (sponge sampling compared to tissue excision).

**Implications**

This study suggests that ESS technology has the potential to greatly reduce the volume of chemical antimicrobial sprays and processing water used in commercial carcass decontamination processes while facilitating uniform carcass coverage. Assuming that relevant pathogen reductions can be achieved using defined antimicrobial chemicals that are food-grade (the focus of future pathogen-inoculated studies at K-State), ESS technology would allow food processing companies to both cut costs on their current antimicrobial treatments by spraying less volume and allow the practical use of more expensive antimicrobials that may provide enhanced antimicrobial effectiveness. This study also showed that an ESS system installed at the Kansas State University Biosecurity Research Institute can be used to inoculate an entire carcass uniformly with target pathogens, including *Salmonella* and Shiga toxin-producing *E. coli*, to support studies to validate the effectiveness of carcass and primal/subprimal antimicrobial intervention technologies, ultimately supporting regulatory approval of such technologies and adoption by meat processors as critical components of integrated meat safety programs.
Figure 1. ESS cabinet dimensions were 6 feet × 5.87 feet × 11.42 feet.
Figure 2. After application of fluorescent dye, the hock area was observed using a black light to determine the uniformity of dye deposition onto all carcass surfaces.

Figure 3. E. coli (log cfu/cm²) recovery at each sample location. Mean E. coli recoveries at locations labeled with different letters are statistically different ($P < 0.05$).