Validation of Antimicrobial Treatments to Reduce *E. coli* O157:H7 and *Salmonella* spp. in Beef Trim and Ground Beef

Project Summary

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Background

Highly publicized outbreaks of food-borne illness since 1993, primarily caused by bacteria such as *E. coli* O157:H7, *Salmonella spp*. and *Listeria monocytogenes*, elicited intense consumer concern about meat safety. In response, regulatory authorities, researchers and the beef industry initiated efforts to implement food safety management systems that would improve microbiological quality. The USDA Food Safety and Inspection Service (FSIS) began initiating new regulatory requirements during the mid-1990s. Packers were required to knife-trim carcasses to remove all visible contaminants, comply with written sanitation standard operating procedures (SSOP), implement Hazard Analysis Critical Control Point (HACCP) systems, and meet microbiological performance criteria and standards for *E. coli* and *Salmonella* as a means to verify HACCP effectiveness and pathogen reduction.

Researchers and beef packers/processors have addressed consumer food safety concerns by developing a variety of methods that are now implemented, or are being further developed, to reduce numbers of bacteria on beef and beef products and improve microbiological safety. These microbiological decontamination technologies include:

- Animal cleaning;
- Chemical dehairing at slaughter;
- Spot-cleaning of carcasses by knife-trimming or steam/hot water vacuuming; and
- Spraying/washing/rinsing of carcasses before evisceration and/or before chilling, with water, chemical solutions and/or steam or hot water.

Ground beef products are commonly implicated as sources for *E. coli* and *Salmonella* outbreaks. While many intervention technologies exist for beef carcasses, very few interventions exist that have been validated for ground beef products. A limited number of studies have been conducted to determine the antimicrobial effects of interventions on beef trim. These results are conflicting and have not been conducted in conditions that simulate a commercial processing facility. The main objective of this study was to validate the effectiveness of acidified sodium chlorite (ASC) and organic acids in reducing pathogen levels in beef trim prior to grinding in a simulated processing environment.

Methodology

Microbiological Analyses

The antimicrobial effects of organic acids and acidified sodium chloride were evaluated by inoculating beef trim, treating it with the intervention and collecting samples at the following points during production:

- Immediately after treatment
- Immediately after grinding
- 24 hours after grinding
- After 5 days of refrigerated storage

- After 30 days of frozen storage

The experiment was conducted in the pathogen processing facility at Texas Tech University under simulated industry conditions.

A total of 240 pounds of beef trim was obtained from the Texas Tech Meat Laboratory. In the pathogen processing area, 200 pounds of the trim was inoculated with either *Salmonella* spp or *E. coli* O157:H7. Two separate sets of trim were prepared with either a high or low level of inoculation. After inoculation (dipping in a pathogen cocktail solution), the trim was held for 20 minutes on sanitized stainless steel tables to allow for pathogen attachment. The control beef trim (40 lbs) was dipped into the sterile water solution and served as the control.

Trim was fed to the grinder using a conveyor belt similar to those used in the industry. Trim was treated by spraying the antimicrobial treatment onto the surface of the trim as it moved down a conveyor towards the grinder. Forty pound portions were sent down the conveyor and treated with one of the 5 treatments: Acidified Sodium Chlorite (1,000 ppm), Lactic Acid-2% or 4%, and Acetic Acid-2% or 4%. The conveyor was cleaned between treatments. Samples of the trim were taken just prior to grinding. The remaining trim was ground and samples were collected just after grinding for microbiological analysis. The remaining ground beef was divided into 3 equal portions, vacuum packaged and two portions were stored at 4^{o}C (39^{o}F) in the processing lab for 5 days. One portion of the ground beef was collected 24 hours after processing and the second 5 days after processing. The other portion of the ground beef was frozen and analyzed 30 days after processing. Control samples were processed before the pathogen inoculated samples and were treated with a water spray instead of an intervention. All experiments were repeated three times. Both *E. coli* and *Salmonella* inhibition were evaluated separately.

Sensory Testing

Preparation of product for sensory testing was similar to the previous experimental design protocol except NONE of the trim was inoculated with pathogens and the sample preparation occurred in the Texas Tech Meat Laboratory. The pathogen processing facility is in the Food Technology building to maintain a physical separation between pathogen inoculated products and consumer products.

Ground products were vacuum packaged and stored at 4^{o}C (39^{o}F) for 5 days or were stored frozen. Sensory tests for comparison of controls and treated samples were conducted on the following time periods:

- Immediately after grinding
- 24 hours after grinding
- 5 days after grinding (refrigerated product)
- 1 month after grinding (after freezing)

Raw products were evaluated for appearance and composition. Beef patties were prepared from the product, cooked to an internal temperature of 71^{o}C (160^{o}F) and evaluated for both flavor and appearance.

Trained sensory panelists (n=24) were given three coded samples, each set of three samples included two of the same samples and one odd sample. Every sample was randomly coded with a three-digit number used for identification purposes. Panelists were asked to taste each sample from left to right and to determine the odd sample.

Findings

Microbiological Analyses

For all samples inoculated with the low dose of *Salmonella* and *E. coli* O157:H7, there were no detectable pathogens after all of the 5 treatments, while the control contained detectable amounts of both of the pathogens. This amount of pathogen load would be more indicative of what would be encountered in the industry, but the actual amount of pathogen reduction is important to determine.

The initial populations of the pathogens on both the *Salmonella* inoculated and *E. coli* inoculated samples (High dose) were not different. Following treatment with all of the 5 treatments, there were significant reductions on the trim prior to grinding for *E. coli* O157:H7 and *Salmonella*. One important observation was that treatment with 4% organic acids did not have additional benefits over the 2% treatments for both *Salmonella* and *E. coli* O157:H7. This is an important economic consideration for processors and also may impact the sensory properties of the product.

The reduction in pathogen loads observed in the trim was sustained during the 5 day refrigerated storage period with the acetic and lactic acid treatments resulting in a 2.5 log cycle reduction in *E. coli* O157:H7 compared to the ground beef prepared from the control trim. ASC treatment resulted in a 1.5 log significant reduction compared to the controls, but was significantly higher than the organic acids. Again, there were no benefits to applying higher concentrations of organic acids. The effects were sustained during the 30 day frozen storage period with all treatments reducing *E. coli* O157:H7 compared to the controls. *Salmonella* reduction was also sustained during refrigerated and frozen storage with the treatments resulting in 1-1.5 log reductions on days 0-5 with no differences among treatments. Samples that were stored frozen had similar reductions in Salmonella that were sustained during freezing.

Sensory Analysis

There was not a significant difference in sensory characteristics determined within or among the five treatments or days of storage. However, at day 30, 4% Acetic Acid and 4% Lactic Acid did show a significant difference at $P<0.10$.

Implications

Ground beef contaminated with *E. coli* and S*almonella* is implicated in many outbreaks of food-borne illness. Interventions at multiple beef processing stages are now in use and treatment of beef trim with acidified sodium chlorite and organic acids could help reduce pathogen levels in ground beef. Higher concentrations of organic acids did not result in additional reduction of the pathogens and therefore the lower concentrations should be used based on economics and sensory properties.