VALIDATION OF PROCESS CAPABILITIES FOR DIRECTLY ACIDIFIED BEEF AND VENISON-CONTAINING BEEF SNACK STICKS FOR CONTROL OF *E. COLI* O157:H7

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Summary

USDA/FSIS guidelines require sausage manufacturers to validate their processes to assure that they can achieve a five-log (99.999%) reduction of *E. coli* O157:H7. Some small meat processors use encapsulated acids instead of lactic acid starter cultures to produce directly acidified sausages. The objectives of this study were to determine 1) the effects of typical thermal processing temperatures and times on reducing *E. coli* O157:H7 in directly acidified all-beef and venison-containing beef snack sticks, 2) the effect of fat content (10 and 25%) on lethality, and 3) the effect of acid type (citric versus lactic) on lethality. For both all-beef and venison-containing beef snack sticks, *E. coli* O157:H7 reductions of approximately 3 log cycles (99.9%) were observed when product internal temperature reached 148 and 155ºF. Reductions increased to more than 5 log cycles after 2 hours of slow drying in which the smokehouse temperature was sequentially decreased to 70°F. Encapsulated citric acid was slightly more effective at lowering product pH, compared with the encapsulated lactic acid. Similar pathogen reductions were observed with 10 and 25% fat content. This study demonstrates that the defined processing schedule used to manufacture beef and venison-containing beef snack sticks is adequate to provide microbiologically safe products and to meet USDA guidelines for pathogen reduction. The processing schedule must include an extended drying phase, in addition to the thermal step, to meet these requirements.

Introduction

In 1994, an *E. coli* O157:H7 outbreak was linked to a dry, fermented, pre-sliced, pork and beef salami product purchased from delicatessen counters in a Seattle grocery chain. Twenty individuals were involved in the salami outbreak, with the median age of 6 years old (range 23 months to 77 years). Three were hospitalized, and one 2-year old developed hemolytic uremic syndrome (HUS), a severe and life-threatening kidney complication. Three individuals in California also became sick from the same incident. Because of this outbreak of *E. coli* O157:H7 being linked to a dry, fermented sausage product for the first time, the USDA/FSIS developed guidelines requiring sausage manufacturers to validate their processes to assure that they can achieve a five-log (99.999%) reduction of *E. coli* O157:H7.

Some small meat processors use encapsulated acids instead of lactic acid starter cultures to produce dry, directly acidified sausages. Encapsulated acids are used to provide the characteristic tangy taste of typically fermented sausage products. Because of the lack of starter cultures for these products, the time required to produce these products is shortened, inasmuch as ripening/fermentation is not required. Directly acidified snack sticks have the potential to cause foodborne illness through *E. coli* O157:H7 because of their low-temperature processing parameters and product properties. This study was designed to quantify the potential for survival of *E. coli*

O157:H7 in beef snack stick products per USDA reduction guidelines.

Procedures

This research project consisted of two phases: Phase 1: all-beef stick validation, and Phase 2: venison-containing beef stick validation. The treatments for each phase were 10 or 25% fat content (green weight) and encapsulated citric or lactic acid. Both control and inoculated batches were prepared. For each phase, a replication consisted of one batch of each treatment being placed in the smokehouse simultaneously. Three replications were completed for each phase.

Fresh beef trimmings and beef fat were obtained from the K-State Meat Lab, and batches were weighed to contain 10% and 25% fat (90% and 75% lean, respectively). The product was ground and formulated with a snack-stick seasoning [Blend 116, Legg's Old Plantation Seasonings, A.C. Legg Inc., Calera, AL] and cure (6.25% sodium nitrite) (A.C. Legg, Packing Co., Inc. Birmingham, AL). The mixture was split into two batches and either encapsulated lactic acid (Meatshure 509, Balchem Encapsulates, Slate Hill, NY) or encapsulated citric acid (Meatshure 333, Balchem Encapsulates) was added. Each batch was then equally divided to provide one non-inoculated batch (control) of each acid and fat content and one inoculated batch of each acid and fat content. For the control batch, 1.2 ounces of sterile deionized water was mixed evenly into the meat batter. The batch receiving the inoculum was spread evenly onto a flat surface in a laminar flow hood to allow for even distribution of inoculum. The inoculum (1.2 ounces of a 5-strain mixture of *E. coli* O157:H7 cultures) was intermittently applied drop-wise over the meat surface and massaged with gloved hands until thoroughly mixed into the meat batter.

The meat batter was stuffed into pre-soaked 0.83-inch-diameter smoked collagen casings (Mid-Western Research & Supply, Wichita, KS). Each stick (18 to 24 inches long) was tied with string, hung vertically on the smokehouse truck, and randomly placed into a commercial smokehouse (Alkar Model 450-UA, Alkar, Lodi, WI). The thermal process included a dry bulb temperature of 110ºF for 1 hour with relative humidity (RH) of 25%, followed by a ripening state at 110ºF and 25% RH for 5 hours. A fast-drying step followed at 120ºF and 25% RH for 40 minutes, followed by 1 hour at 130ºF and 25% RH, 40 minutes at 140ºF and 25% RH, and a hot-air finish with the smokehouse temperature at 180ºF and 25% RH until the internal product temperature reached 155ºF. A cool down slow drying period followed for 20 minutes at 130ºF, 20 minutes at 110ºF, 20 minutes at 90ºF, and 1 hour at 70ºF. The pH was measured in duplicate on both control and inoculated samples.

***E. coli* O157:H7 Enumeration**. Raw meat control samples were collected before inoculation and also were collected from the control batches to assure that no pathogenic *E. coli* O157:H7 was in the meat before inoculation. Raw inoculated samples were taken from the inoculated meat batter to determine the initial inoculum content of the product. Both selective and injury recovery media were used for the enumeration *E. coli* O157:H7 populations in each sample.

Heat-treated samples (one inoculated and one non-inoculated control) were collected when the internal product temperature reached 148ºF and 155ºF, as well as at the end of the drying cycle. The non-inoculated control sample was used for proximate analysis, pH, titratable acidity, and water activity. The inoculated sample was used for *E. coli* O157:H7 enumeration and pH analysis. Samples and sterile diluent were blended, and serial dilutions were plated on agar.

***E. coli* O157:H7 Enrichment**. Modified *E. coli* broth containing sodium novobiocin

antibiotic was used for enrichment of heat-treated sausage samples. The samples were incubated in modified *E. coli* broth overnight, followed by streaking onto selective agar plates. This enrichment procedure was used to detect very low counts of surviving organisms and injured cells that would not normally be detected by direct sample plating.

Results and Discussion

The *E. coli* O157:H7 inoculation rates for meat batters were approximately 12 and 10 million colony forming units per gram for all-beef and venison-containing beef, respectively. Smokehouse heating to 148 and 155°F internal sausage temperature resulted in 99.8% and 99.9% reductions in *E. coli* O157:H7, respectively, for all-beef sticks. Similar reductions for venison-containing beef sticks were 99.3% for both heating temperatures. These reductions (99.9%, or 3 log reductions) would not meet the five-log (99.999%) USDA/FSIS pathogen control guidelines for this product group. After the specified drying process for the products had been completed, however, counts had been further reduced to very low surviving numbers, or not detected at all. Therefore, the complete manufacturing process (acidification, thermal processing, and drying) for this product did meet USDA/FSIS guidelines.

The pH of both control and inoculated samples for all-beef and venison-containing beef snacks were very similar for all treatments and sampling times. The final pH range for all products at the end of the drying cycle was 4.7 to 5.3. When comparing between citric and lactic acid treatments, however, pH was higher for products with lactic acid. In addition, the venison-containing beef snack sticks demonstrated higher pH than the all-beef snack sticks did. Similar bacterial reductions were observed for treatments having differing fat contents (10 vs. 25%).