Food Safety Systems, LLC

Research Report

Treatment of Inoculated Sliced Turkey Breast for Control of *Listeria monocytogenes* using AMS Surface and Air Sanitation Technology

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Introduction:

Ready-to-eat (RTE) meats contaminated with *L. monocytogenes* during processing are an important public health threat and one that has received substantial industry and governmental attention for over two decades. Recently, the USDA issued stringent policy guidelines to govern the production of RTE meat products to ensure minimal risk for *L. monocytogenes* contamination through mandated microbiological testing of products and the processing environment, along with the use of validated antimicrobial intervention treatments to reduce contamination levels on products and prevent *Listeria* outgrowth during storage. The necessity for interventions that can reduce and control this biological hazard is a priority for RTE meat processors.

The objective of this study was to determine the efficacy of the AMS treatment for control of *Listeria monocytogenes* on inoculated sliced turkey breasts. This technology is considered a processing aid and does not require labeling. It is also a dry process and does not involve the addition of water in the form of chemical sprays. It is designed to be scalable and easily implemented into a RTE plant of any size. This system is applicable to the production of RTE meats for the reduction and control of *L. monocytogenes* contamination of the product surfaces and surrounding contact surfaces.

Materials and Methods

Vacuum packaged sliced turkey breast product was provided by a USDA Inspected Turkey Processor for this study. The product was stored in a 34°F cooler until it was removed from the packaged and prepared for inoculation and treatment.

Cultures and Product Inoculation:

The following cultures were utilized: *Listeria monocytogenes;* Scott A-2, V7-2, 4B, 1/2a, 1/2b (ABC Research Corporation, Gainesville, FL). The stock cultures were cultivated by placing one impregnated bead into a 5 ml solution of Difco® Tryptic Soy Broth (TSB) and incubated for 24 h at 35°C. Next, a 0.05 ml loop of the respective culture was used to inoculate a 5 ml solution of TSB, with incubation for 24 h at 35°C. After incubation, 1 ml of the respective culture was used to inoculate 49 ml TSB incubated for 24 h at 35°C. Following incubation, samples were centrifuged (15,300 X g at 4°C), the supernatant decanted and the pellet resuspended with 50 ml of 0.1% peptone and centrifuged (15,300Xg at 4°C) a final time. The peptone was decanted and the remaining pellet resuspended with 10 ml of 0.1% peptone. The five 10 ml bottles of respective culture was mixed together to create a 50 ml mixed-strain cocktail containing 10^9 to 10^{10}CFU/ml of *Listeria monocytogenes.* Cultures were confirmed by cultivation on selective and differential media, and biochemical analysis of presumptive colonies using API-Listeria kits.

The slices of turkey breasts were inoculated on the exterior surfaces with a five-strain cocktail of *L. monocytogenes* inoculum. The inoculum was misted onto the tissue surfaces using a plastic spray bottle with samples contained within a sealed inoculation chamber. The inoculum was allowed 30 minutes for attachment to the meat surfaces. Immediately prior to treatment applications, the surfaces of the inoculated products were sampled and analyzed to establish the actual inoculum level of attached *L. monocytogenes*. The inoculation level for the single replications was 6.2 log cfu/cm^2.

Application of AMS Treatment:

The inoculated slices of turkey breasts were treated in a controlled environmental chamber equipped with an AMS Surface and Air Sanitation System for periods of 0, 15 seconds, 30 seconds and 60 seconds. The temperature in the controlled environmental chamber was 73° F and the relative humidity was 55%. The target surface inoculation was 6.0 Log CFU/cm^2. After each treatment, the inoculated turkey breast slices were tested to determine levels of Lm. The study was conducted at 10°C in the Kansas State University Food Safety Processing Laboratory.

A negative control was conducted in which inoculated samples were placed in a controlled environmental chamber without the AMS system. Samples were removed from the chamber at 0, 15, 30 and 60 seconds.

Sampling and Chemical and Microbial Analysis:

Turkey breasts slices were collected and placed in a sterile bag after treatment. The samples were diluted with 90 ml of 0.1% sterile peptone water (PW) and homogenized in a stomacher for one minute. Samples were serially diluted in sterile PW and spiral plated onto Modified Oxford Agar (MOX; Oxoid ltd., Basingstoke, Hampshire, England) using a Whitley automatic spiral plater. The plates were then incubated at 35 °C for 24 h and enumerated.

Non-inoculated Turkey breasts slices were treated using the AMS Surface and Air Sanitation System for periods of 0, 15, 30 and 60 seconds and tested for residual levels of ozone and hydrogen peroxide using gas spectrometry. TBA values were also measured (TBAR assay) on treated Turkey breasts slices.

Results and Discussion:

Results from this preliminary study can be found in Table 1. Log CFU/cm^2 reductions were calculated as the difference in log recoveries from the inoculated products prior to treatment and the log recovery after treatment.

Table 1. Average recoveries (Log CFU/cm^2) of *Listeria monocytogenes* on inoculated turkey breast slices treated in the Controlled Environmental Chamber equipped with AMS Surface and Air Sanitation Technology for periods of 0, 15, 30 and 60 seconds v. Control (untreated samples).

Sample	Treated Samples *Listeria monocytogenes*	Control Samples *Listeria monocytogenes*
0 time	**6.20**	**6.20**
15 Seconds	**5.10**	**6.20**
30 Seconds	**4.50**	**6.10**
60 Seconds	**4.10**	**6.15**

This study demonstrates the efficacy of the AMS Surface and Air Sanitation Technology for the surface decontamination of sliced turkey breast inoculated with *Listeria monocytogenes*. Reductions greater than 1.0 log CFU/cm^2 were observed after the 15 second treatment. The 30 second treatment resulted in a reductions of 1.7 log CFU/ cm^2. The 60 second treatment resulted in a 2.1 log CFU/cm^2 reduction. Reductions in the negative control were less than 0.1 log CFU/cm^2 for each sampling time (0, 15, 30 and 60 seconds).

Residue testing showed no detectible residual levels of ozone or hydrogen peroxide in sliced turkey samples. TBA values were similar between treated and untreated samples, 0.16 and 0.14, respectively.

During the course of the study ambient ozone levels were measured using a Model 500 Aeroqual (New Zealand) monitoring instrument. Ozone levels were recorded at 0.038 PPM inside the controlled environmental chamber. Hydrogen peroxide levels were measured using Dragger tubes. The levels for hydrogen peroxide ranged from 0.115 – 0.135 ppm.