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# Investigating the Control of *Listeria monocytogenes* on a Ready-to-Eat Ham Product Using Natural Antimicrobial Ingredients and Postlethality Interventions

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## Abstract

Ready-to-eat (RTE) meat and poultry products manufactured with natural or organic methods are at greater risk for *Listeria monocytogenes* growth, if contaminated, than their conventional counterparts due to the required absence of preservatives and antimicrobials. Thus, the objective of this study was to investigate the use of commercially available natural antimicrobials and postlethality interventions in the control of *L. monocytogenes* growth and recovery on a RTE ham product. Antimicrobials evaluated were cranberry powder (90MX), vinegar (DV), and vinegar/lemon juice concentrate (LV1X). Postlethality interventions studied were high hydrostatic pressure at 400 (HHP400) or 600 (HHP600) MPa, lauric arginate (LAE), octanoic acid (OA), and postpackaging thermal treatment (PPTT). Parameters evaluated through 98 days of storage at $4 \pm 1°C$ were residual nitrite concentrations, pH, $a_w$, and viable *L. monocytogenes* on modified Oxford (MOX) media. On day 1, OA, 90MX, DV, and LV1X yielded lower residual nitrite concentrations than the control, whereas HHP400, HHP600, and LAE did not. LAE, HHP400, and OA reduced *L. monocytogenes* population compared to the control after 1 day of storage by 2.38, 2.21, and 1.73 $\log_{10}$ colony-forming units per gram, respectively. PPTT did not achieve a significant reduction in *L. monocytogenes* populations. *L. monocytogenes* recovered and grew in all postlethality intervention treatments except HHP600. 90MX did not inhibit the growth of *L. monocytogenes*, while DV and LV1X did. Results of this study demonstrate the bactericidal properties of HHP, OA, and LAE and the bacteriostatic potential of natural antimicrobial ingredients such as DV and LV1X against *L. monocytogenes*.

## Introduction

Regulations that govern natural and organic foods do not permit the direct addition of nitrite or nitrate, curing ingredients typically used for cured meat products. Consequently, use of celery (*Apium graveolens* var. dulce) as a natural source of nitrate has become common in the meat industry for producing natural and organic processed meats with cured meat properties (Sebranek and Bacus, 2007; Sindelar and Milkowski, 2011).

*Listeria monocytogenes* is a foodborne pathogen of concern (CFR, 2003). Ready-to-eat (RTE) meat and poultry products have been associated with listeriosis outbreaks and, consequently, factors affecting growth of *L. monocytogenes* in such products have received considerable attention.

RTE meat and poultry products manufactured with natural or organic methods and labeled as such as required by regulation (USDA FSIS, 2005) are at increased risk for *L. monocytogenes* growth if contaminated (Schrader, 2010; Sullivan, 2011, Sebranek *et al.*, 2012; Sullivan *et al.*, 2012a, 2012b).

The use of postlethality interventions for control of *L. monocytogenes* in RTE natural or organic meat and poultry products is of interest because some of these technologies are allowed for these products. For example, high hydrostatic pressure processing (HHP) is a postlethality intervention that is acceptable for natural and organic products (USDA FSIS, 2012). Other examples of acceptable postlethality interventions include sprays or solutions such as lauric arginate (lauramide arginine ethyl ester or LAE) and octanoic acid (sometimes referred to as caprylic acid or OA) as well as postpackaging thermal pasteurization.

Consequently, investigating commercially available natural antimicrobial ingredients and postlethality interventions currently allowed for meat and poultry products labeled as

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natural or organic to inhibit *L. monocytogenes* was the objective of this study.

## Materials and Methods

### *Manufacture of hams*

Nine ham formulations (eight experimental and one control) were manufactured for comparison of individual treatments; no treatment combinations were included. Hams were produced at the Iowa State University Meat Laboratory using 18.14 kg of ham insides, 3.66 kg water, 0.50 kg salt, 0.30 kg sugar, and 74.84 g celery powder plus the selected antimicrobials or postlethality interventions. The ham muscles were obtained from a local processor, coarse ground (9.53-mm hole plate) (Biro Manufacturing Co., Marblehead, OH) and mixed with nonmeat ingredients for 2 min (Leland Southwest, Fort Worth, TX). All products were formulated to contain 50 mg/kg natural nitrite from celery powder (VegStable 504, Florida Food Products, Inc., Eustis, FL). Mixed samples were then reground (6.35-mm-diameter hole plate) and stuffed into a 50-mm-diameter impermeable plastic casing (Nalobar APM 45, Kalle USA, Gurnee, IL). All treatments were then placed in a smokehouse (Maurer, AG, Reichenau, Germany), heated to 71.1°C internally, and placed in a 0°C cooler to stabilize. The next day, the hams were sliced into 12.0-mm-thick slices (SE 12 D, Bizerba, Piscataway, NJ), placed into barrier bags (B2470, Cryovac Sealed Air Corporation, Duncan, SC; oxygen transmission rate of 3–6 cc/$m^2$ at 4°C (0% RH, 24 h) and water vapor transmission rate of 0.5–0.6 g/0.6 $m^2$ at 38°C (100% RH, 24 h) as single slices in each bag, and vacuum sealed (UV 2100, Multivac, Inc., Kansas City, MO). Hams for chemical analyses were stored at 4 ± 1°C. Hams for microbial analyses were transferred to the Iowa State University Microbial Food Safety Laboratory for immediate inoculation, then stored at 4 ± 1°C. Two independent replications were produced.

Three natural antimicrobial ingredients were evaluated; cranberry powder (90MX; Ocean Spray International, Middleboro, MA), vinegar (DV; WTI Ingredients, Inc., Jefferson, GA), and vinegar/lemon juice concentrate (LV1X; WTI Ingredients, Inc., Jefferson, GA) (wt/wt). Each ingredient was added to the hams at a concentration (1.0%, 1.0%, and 2.5%, respectively) recommended by the supplier.

On day 0, five randomly selected slices of ham from the control, 90MX, DV, and LV1X formulations were weighed and measured ($n = 20$ per replication) to obtain representative weight and surface area measurements, which were then used to calculate $\log_{10}$ colony-forming units (CFU) per square centimeter, and OA and LAE volumes per slice to be used in the study, respectively.

### Analytical Measurements

Proximate analysis was conducted on duplicate samples for fat, moisture, and protein on all formulations on day 0 using Association of Official Analytical Chemists (AOAC) methods 960.63, 950.46, and 992.15, respectively (AOAC, 1990a, 1990b, 1993).

Product pH was measured on duplicate samples by placing a pH probe (FC20, Hanna Instruments, Woonsocket, RI) into homogenized (KFP715 food processor, Kitchenaid, St. Joseph, MI) ham samples (Sebranek and others, 2001), using a pH/ion meter (Accumet 925 pH/ion meter, Fisher Scientific).

Available moisture ($A_w$) was determined on duplicate samples of all formulations using a water activity meter (AquaLab 4TE, Decagon Devices Inc., Pullman, WA).

Residual nitrite was determined utilizing AOAC method 973.31 (AOAC, 1990c) for duplicate samples.

### *Preparation of inoculum and sample inoculation*

*L. monocytogenes* strains Scott A NADC 2045 serotype 4b, H7969 serotype 4b, H7962 serotype 4b, H7596 serotype 4b, and H7762 serotype 4b were obtained from the Iowa State University Microbial Food Safety Laboratory. Each strain was cultured in tryptic soy broth supplemented with 0.6% yeast extract (TSBYE; Difco, Becton Dickinson, Sparks, MD) for 24 h at 35°C. A minimum of two consecutive 24-h transfers of each strain to fresh TSBYE (35°C) were performed. The bacterial cells from all strains were harvested by centrifugation (10 min at 10,000 rpm and 4°C) in a Sorvall Super T21 centrifuge (American Laboratory Trading, Inc., East Lyme, CT), and the pelleted cells were resuspended together to form a 5-strain mixed culture in 30.0 mL of sterile buffered peptone water (BPW; Difco, Becton Dickinson). The concentration of the mixed culture was approximately $10^9$ CFU/mL based on plate counts using TSBYE agar. Two serial dilutions (100-fold each) of the cell suspension were prepared in BPW to give a final inoculum concentration of $10^5$ CFU/mL to be used for ham inoculations.

For inoculation, each packaged sample was reopened and the product surface aseptically inoculated with a 0.2-mL aliquot of the diluted five-strain mixed culture. The cell concentration at inoculation was approximately $10^3$ CFU/g ($10^{1.4}$ per $cm^2$) of ham slice. The bags were then vacuum sealed and stored at 4 ± 1°C.

### *Postlethality interventions*

Four postlethality interventions were evaluated: HHP, OA, LAE, and postpackaging thermal treatment (PPTT). All postlethality interventions were applied to the products, including controls, within 2 h after inoculation; consequently there was no concern for biofilm formation by *L. monocytogenes* as reported by several authors (Gandhi and Chikindas, 2007; Desai and others, 2012).

HHP was evaluated at two pressure levels: 400 MPa, 4-min dwell time at 12 ± 2°C (initial temperature of the pressurization fluid) or 600 MPa, 4-min dwell time at 12 ± 2°C, with 600 MPa the commercial standard. Inoculated hams were subjected to the appropriate HHP treatment using a FOODLAB 900 Plunger Press system (Standsted Fluid Power Ltd., Standsted, UK). The pressurization fluid was a 50.0% propylene glycol (GWT Koilguard; GWT Global Water Technology, Inc., Indianapolis, IN) and 50.0% water solution (vol/vol). The average rate of pressurization was 350 MPa/min and depressurization occurred within 7 s. Adiabatic heating of the pressurization fluid was 4.6 ± 0.8 °C /100 MPa.

A 23.4% Octa-Gone solution (vol/vol) (Octa-Gone; Eco-Lab, Inc., Eagan, MN) was prepared by mixing OA with sterile de-ionized water at 4 ± 1°C. Based on average surface area measurements, the OA solution was aseptically dispensed into the bag containing the ham slice (0.0186 mL/$cm^2$) and vacuum sealed.

A 2.5% Protect-M (Protect-M; Purac America, Lincolnshire, IL) and 97.5% water solution (vol/vol) was prepared by

Table 1. Effect of Natural Antimicrobial Ingredients on Physicochemical Properties of Alternatively Cured Ready-to-Eat Ham[a]

| *Treatment*[b] | $a_w$ | *pH* | *Fat %* | *Moisture %* | *Protein %* |
|---|---|---|---|---|---|
| Control | $0.9745^{A}$ | $6.14^{BC}$ | 1.95 | $76.29^{B}$ | 17.85 |
| 90MX | $0.9736^{B}$ | $5.85^{A}$ | 1.85 | $75.60^{AB}$ | 18.04 |
| DV | $0.9695^{C}$ | $6.04^{CD}$ | 2.04 | $75.49^{A}$ | 18.20 |
| LV1X | $0.9693^{C}$ | $6.03^{D}$ | 1.89 | $75.78^{AB}$ | 18.09 |
| SE[c] | 0.0002 | 0.04 | 0.12 | 0.24 | 0.22 |

[a]Values are least-squares means. Within a column, means with different superscripts (A through D) are significantly different ($p < 0.05$).
[b]Control, alternatively cured control; 90MX, cranberry powder; DV, vinegar; LV1X, vinegar and lemon juice concentrate.
[c]Standard error of the differences of least-squares mean.

mixing Protect-M (LAE) with sterile de-ionized water at 4 ± 1°C, aseptically dispensed into the bag containing the ham slice (0.007192 mL/cm$^2$) and vacuum sealed.

PPTT was conducted by immersing packages of ham in water at 71.0 ± 1.0°C for 30 s using a water bath (Isotemp-228, Fisher Scientific), based on results reported by Chen and others (Chen *et al*., 2004). Packages were held in heated water for 30 s, placed on ice to chill, then stored at 4 ± 1°C.

### *Microbial analysis*

Microbial analysis of ham samples for viable *L. monocytogenes* was conducted on days 1, 14, 28, 42, 56, 70, 84, and 98. On the appropriate day, two packages for each treatment were opened aseptically, and their contents were placed inside a sterile Whirl-Pak stomacher bag (Nasco, Ft. Atkinson, WI). Fifty milliliters of sterile BPW were added and the bags were shaken by hand for approximately 30 s. The rinse solution was then serially diluted (10-fold) in BPW. Either 1.0 or 0.1 mL of the appropriate dilution was surface-plated on modified *Listeria*-selective agar (Oxford, MOX; Difco, Becton Dickinson). Agar plates were incubated at 35°C for 48 h, after which colonies typical of *L. monocytogenes* were enumerated. The populations (CFU/mL) were averaged and then converted to $\log_{10}$ CFU/cm$^2$ using the average area of the ham slices ($n = 40$). The detection limit of our sampling protocols was $\geq 0.30$ $\log_{10}$ CFU/g based on a sample weight of 25.0 g.

### *Statistical analysis*

The overall design of the experiment was a factorial design. The generalized linear mixed models (GLIMMIX) procedure of Statistical Analysis System (version 9.3, SAS Institute Inc., Cary, NC) was used for statistical analysis. All data were analyzed for treatment effects within day. Day and treatment × day interactions were also analyzed. Where significant effects ($p < 0.05$) were found, pairwise comparisons between the least squares means were computed for each day using Tukey's HSD adjustment.

## Results and Discussion

### *Mean surface area and weight results*

The mean weight of the ham slices was 25.24 ± 0.58 g, while the mean surface area was 53.68 ± 1.44 cm$^2$, respectively (data not shown; $n = 40$). These dimensions resulted in LAE and OA treatment volumes of 0.39 and 1.00 mL per package, and concentrations of 38.24 and 333.27 mg/kg, respectively.

### *Physicochemical traits of hams*

The LV1X and DV treatments resulted in significantly lower $a_w$ values (Table 1) than the 90MX and control treatments ($p < 0.05$). The 90MX treatment, in turn, resulted in

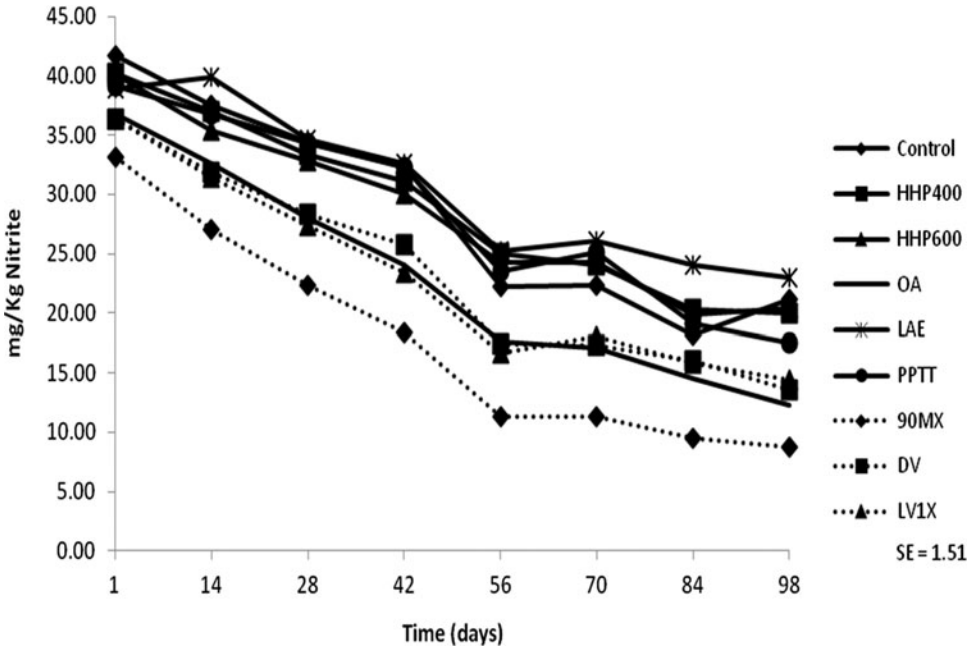


**FIG. 1.** Effect of treatment on residual nitrite concentration of alternatively cured ready-to-eat ham stored at 4 ± 1°C.

significantly lower $a_w$ values than the control ($p < 0.05$). Final product pH was also affected by the antimicrobial added. The pH of the control was not different from that of the DV treatment ($p > 0.05$), but differed from both the LV1X and 90MX treatments ($p < 0.05$). Cranberry contains phenolic acids with a high titratable acidity (Lee *et al.*, 2006), and vinegar and vinegar/lemon juice concentrates are also reservoirs of acidic compounds, such as acetic and citric acid.

### *Residual nitrite concentration*

Treatment and day exerted a significant effect (Fig. 1) on residual nitrite ($p < 0.05$). Additionally, a significant treatment × day interaction was observed ($p < 0.05$). Although all ham formulations were manufactured with 50 mg/kg natural nitrite, the highest residual nitrite concentration observed on day 1 of the study was 41.67 mg/kg, indicating that part of the nitrite was depleted in curing reactions that took place during product manufacture (Honikel, 2008). On day 1 of the study, the OA, 90MX, DV, and LV1X treatments exhibited lower residual nitrite concentrations than the control ($p < 0.05$), probably due to the acidic nature of the natural antimicrobials.

### *Bacteriological results*

Treatment and day had a significant ($p < 0.05$) effect on viable *L. monocytogenes* populations (Fig. 2). However, the natural antimicrobials did not affect *L. monocytogenes* populations after 1 day of storage ($p > 0.05$) when compared to the control treatment.

The DV treatment did not show changes in *L. monocytogenes* populations throughout the study ($p > 0.05$), while the LV1X treatment did not exhibit increased *L. monocytogenes* populations until day 84 ($p < 0.05$). *L. monocytogenes populations* found in the LV1X treatment on days 84 and 98, however, were still lower than those of the control ($p < 0.05$). These results suggest that, at the levels used in this study, DV and LV1X exhibited bacteriostatic properties against *L. monocytogenes*.

The HHP600 treatment resulted in *L. monocytogenes* populations below the detection limit of our sampling protocols (≥0.30 $\log_{10}$ CFU/g) throughout the study. These results agree with those obtained by Myers and others (2013a, 2013b), who found that an HHP treatment of 600 MPa for 3 min and 17°C resulted in a 3.85–4.35 $\log_{10}$ CFU/g reduction in *L. monocytogenes* populations on RTE meat products. The LAE, HHP400, and OA treatments resulted in 2.38, 2.21, and 1.73 $\log_{10}$ CFU/g reductions in viable *L. monocytogenes* populations, respectively, compared to the control, on day 1.

The PPTT did not significantly decrease initial viable *L. monocytogenes* populations ($p > 0.05$) compared to the control. These results contrast to those by Chen and others (2004), who observed that a postpackaging thermal treatment of 71 ± 1°C for 30 s resulted in a 1.4 $\log_{10}$ CFU/g reduction in *L. monocytogenes* populations on frankfurters when using a 3.4 $\log_{10}$ CFU/g initial inoculation. It is not clear why our results differed from those reported by Chen and others (2004).

Although initial bactericidal effects of the HHP400, OA, and LAE treatments were observed, bacteriostatic properties of these treatments were limited. The HHP400 treatment resulted in an ($p < 0.05$) increase in viable *L. monocytogenes* populations after 56 days. In fact, by day 98, there was no difference between the control and the HHP400 treatments ($p > 0.05$).

Similarly, the OA and LAE treatments showed significant ($p < 0.05$) increases in *L. monocytogenes* populations by days 28 and 14, respectively, with both of these treatments showing no difference compared to the control by day 70 ($p > 0.05$). The LAE treatment, in fact, resulted in viable *L. monocytogenes* populations that were not different from those found in the control as early as day 28 ($p > 0.05$). These findings agree with

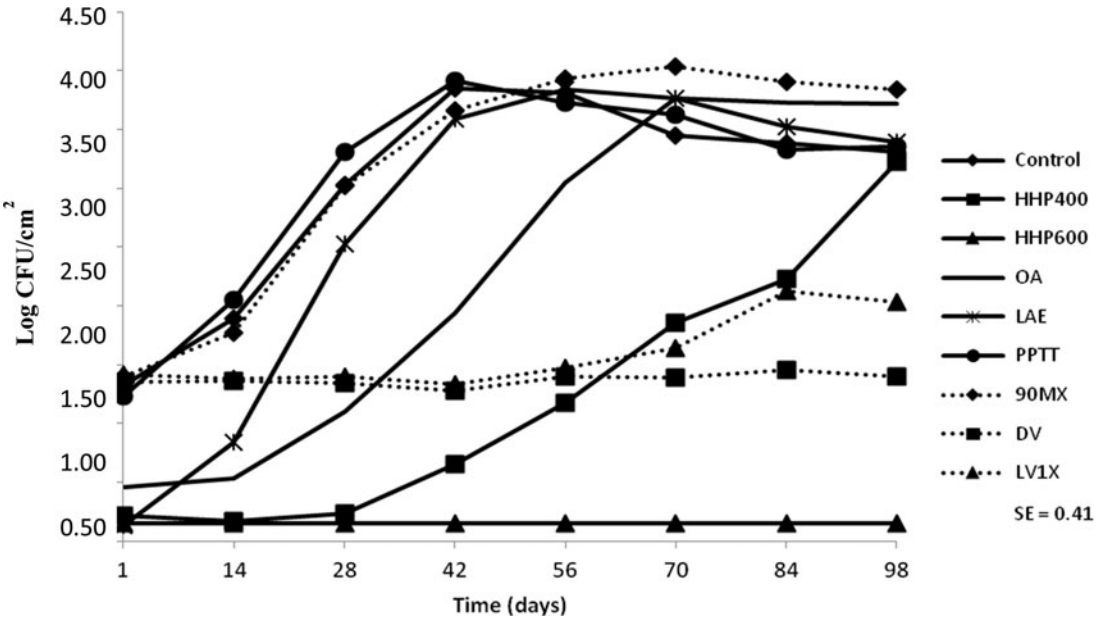


**FIG. 2.** Effect of treatment on a five-strain mixed culture of viable *Listeria monocytogenes* (log colony-forming units (CFU)/cm$^2$) on modified Oxford medium on alternatively cured ready-to-eat ham stored at 4 ± 1°C.

Porto-Fett and others (2010), who reported that only in combination with lactate or diacetate did lauric arginate exert a bacteriostatic effect. Thus, although they may provide initial lethality, lauric arginate and OA alone do not inhibit any *L. monocytogenes* that may survive. Our study indicates that, while beneficial for initial lethality, HHP400, OA, and LAE did not offer protection against the growth of surviving *L. monocytogenes* during storage.

## Conclusions

At the levels used and under the conditions of this study, DV and LV1X exhibited bacteriostatic properties against *L. monocytogenes* and represent viable options for manufacturers of organic and natural processed meat and poultry products, but did not exhibit bactericidal properties. Although beneficial for initial lethality, the HHP400, OA, and LAE postlethality interventions did not prevent growth of surviving *L. monocytogenes* during storage. The HHP600 treatment, however, which is currently used by the industry, was very effective. Thus, additional research on combining natural antimicrobial ingredients and postlethality interventions that are not fully effective by themselves as well as assessing the organoleptic effects of the added ingredients used as antimicrobials in the manufacture of organic and natural processed meat and poultry products is warranted.

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## Disclosure Statement

No competing financial interests exist.

## References

[AOAC] Association of Official Analytical Chemists International. Fat (crude) or ether extract in meat. Official method 960.39. In: *Official Methods of Analysis*. Arlington, VA: AOAC International, 1990a.

AOAC. Moisture in meat. Official method 950.46. In: *Official Methods of Analysis*. Arlington, VA: AOAC International, 1990b.

AOAC. Nitrites in Cured Meat. Official Method 973.31. In: *Official Methods of Analysis*. Arlington, VA: AOAC International, 1990c.

AOAC. Crude protein in meat and meat products. Official method 992.15. In: *Official Methods of Analysis*. Arlington, VA: AOAC International, 1993.

Chen CM, Sebranek JG, Dickson JS, Mendonca AF. Combining pediocin (ALTA 2341) with postpackaging thermal pasteurization for control of *Listeria monocytogenes* on frankfurters. J Food Prot 2004;67:1855–1865.

[CFR] Code of Federal Regulations. 9 CFR 430. Control of *Listeria monocytogenes* in post-lethality exposed ready-to-eat products; final rule. 2003. Available at: http://www.fsis.usda.gov/OPPDE/rdad/FRPubs/97-013F/Lm_Rule_Compliance_Guidelines_2004.pdf, accessed July 4, 2013.

Desai MA, Soni KA, Nannapaneni R, Schilling MW, Silva JL. Reduction of *Listeria monocytogenes* biofilms on stainless steel and polystyrene surfaces by essential oils. J Food Prot 2012;75:1332–1337.

Gandhi M, Chikindas ML. Listeria: A foodborne pathogen that knows how to survive. Int J Food Micro 2007;113:1–15.

Honikel K-O. The use and control of nitrate and nitrite for the processing of meat products. Meat Sci 2008;78:68–76.

Lee C, Reed JD, Richards MP. Ability of various polyphenolic classes from cranberry to inhibit lipid oxidation in mechanically separated turkey and cooked ground pork. J Muscle Foods 2006;17:248–266.

Myers KL, Cannon J, Montoya D, Dickson JS, Lonergan S, Sebranek JG. Effects of high hydrostatic pressure and varying concentrations of sodium nitrite from traditional and vegetable-based sources on the growth of *Listeria monocytogenes* on ready-to-eat (RTE) sliced ham. Meat Sci 2013a;94:69–76.

Myers KL, Montoya D, Cannon J, Dickson JS, Sebranek JG. The effect of high hydrostatic pressure, sodium nitrite and salt concentration on the growth of *Listeria monocytogenes* on RTE ham and turkey. Meat Sci 2013b;93:263–268.

Porto-Fett ACS, Campano SG, Smith JL, Oser A, Shoyer B, Call JE, Luchansky JB. Control of *Listeria monocytogenes* on commercially-produced frankfurters prepared with and without potassium lactate and sodium diacetate and surface treated with lauric arginate using the Sprayed Lethality in Container (SLIC®) delivery method. Meat Sci 2010;85:312–318.

Schrader KD. Investigating the control of *Listeria monocytogenes* on uncured, no-nitrate- or-nitrite-added meat products [PhD dissertation]. Ames, IA: Iowa State University, 2010.

Sebranek JG, Bacus JN. Cured meat products without direct addition of nitrate or nitrite: What are the issues? Meat Sci 2007;77:136–147.

Sebranek JG, Jackson-Davis AL, Myers KL, Lavieri NA. Beyond celery and starter culture: Advances in natural/organic curing processes in the United States. Meat Sci 2012;92:267–273.

Sebranek JG, Lonergan SM, King-Brink MA, Larson EL, Beermann DH. *Meat Science and Processing,* 3rd ed. Zenda, WI: Peerage Press, 2001.

Sindelar JJ, Milkowski AL. Sodium nitrite in processed meat and poultry meats: A review of curing and examining the risk/benefit of its use. White Paper Series, Number 3. Champaign, IL: American Meat Science Association, 2011.

Sullivan GA. Naturally cured meats: Quality, safety, and chemistry [PhD dissertation]. Ames, IA: Iowa State University, 2011.

Sullivan GA, Jackson-Davis AL, Niebuhr SE, Xi Y, Schrader KD, Sebranek JG, Dickson JS. Inhibition of *Listeria monocytogenes* using natural antimicrobials in no-nitrate-or-nitrite-added ham. J Food Prot 2012a;75:1071–1076.

Sullivan GA, Jackson-Davis AL, Schrader KD, Xi Y, Kulchaiyawat C, Sebranek JG, Dickson JS. Survey of naturally and conventionally cured commercial frankfurters, ham, and bacon for physio-chemical characteristics that affect bacterial growth. Meat Sci 2012b;92:808–815.

[USDA FSIS] U.S. Department of Agriculture Food Safety Inspection Services. Food standards and labeling policy book. 2005. Available at: http://www.fsis.usda.gov/OPPDE/larc/

Policies/Labeling_Policy_Book_082005.pdf, accessed January 2, 2014.

USDA FSIS. FSIS compliance guideline: Controlling *Listeria monocytogenes* in post-lethality exposed ready-to-eat meat and poultry products. 2012. Available at: http://www.fsis.usda.gov/shared/PDF/Controlling_LM_RTE_guideline_0912.pdf, accessed July 4, 2013.

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