Journal of Food Protection, Vol. 76, No. 7, 2013, Pages 1245–1249
doi:10.4315/0362-028X.JFP-12-528

Research Note

Determination of 5-Log Reduction Times for *Escherichia coli* O157:H7, *Salmonella enterica,* or *Listeria monocytogenes* in Acidified Foods with pH 3.5 or 3.8†

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MS 12-528: Received 26 November 2012/Accepted 22 February 2013

ABSTRACT

A critical factor in ensuring the safety of acidified foods is the establishment of a thermal process that assures the destruction of acid-resistant vegetative pathogenic and spoilage bacteria. For acidified foods such as dressings and mayonnaises with pH values of 3.5 or higher, the high water phase acidity (acetic acid of 1.5 to 2.5% or higher) can contribute to lethality, but there is a lack of data showing how the use of common ingredients such as acetic acid and preservatives, alone or in combination, can result in a 5-log reduction for strains of *Escherichia coli* O157:H7, *Salmonella enterica,* and *Listeria monocytogenes* in the absence of a postpackaging pasteurization step. In this study, we determined the times needed at 10°C to achieve a 5-log reduction of *E. coli* O157:H7, *S. enterica,* and *L. monocytogenes* in pickling brines with a variety of acetic and benzoic acid combinations at pH 3.5 and 3.8. Evaluation of 15 different acid-pH combinations confirmed that strains of *E. coli* O157:H7 were significantly more acid resistant than strains of *S. enterica* and *L. monocytogenes*. Among the acid conditions tested, holding times of 4 days or less could achieve a 5-log reduction for vegetative pathogens at pH 3.5 with 2.5% acetic acid or at pH 3.8 with 2.5% acetic acid containing 0.1% benzoic acid. These data indicate the efficacy of benzoic acid for reducing the time necessary to achieve a 5-log reduction in target pathogens and may be useful for supporting process filings and the determination of critical controls for the manufacture of acidified foods.

The safe production of acidified foods depends on maintaining a pH below 4.6 to prevent spore germination and outgrowth of *Clostridium botulinum* as mandated in the Code of Federal Regulations (21 CFR 114) *(20)*. This same regulation also requires that vegetative cells of microorganisms of public health significance be destroyed during the processing and manufacture of acidified foods. Destruction of bacterial pathogens is typically achieved by a heat process *(2, 3)*. However, recent data indicate that if a pH of 3.3 or below is maintained, acidified foods can be rendered safe without a heat treatment, assuming acetic acid is the primary acidulent *(1)*. To assure a 5-log reduction of *Escherichia coli* O157:H7, *Salmonella enterica,* and *Listeria monocytogenes,* products must be held before sale and distribution for approximately 2 days at or above 25°C or 6 days at or above 10°C, allowing time for the acetic acid to kill the pathogens. The conditions needed for achieving a 5-log reduction of pathogens in acidified foods at higher pH values or with specific acetic acid concentrations have not been defined. Acidified food products that cannot meet the pH 3.3 requirement or be heat processed without significantly altering sensory properties include dressings, mayonnaises, and some pickled vegetable products. A 5-log reduction of vegetative pathogens in these products may be defined as a critical control point for safe production of these products under the Food Safety Modernization Act (FSMA) *(19)*.

Acidified foods such as dressings often contain mustard, horseradish, peppers, or other compounds that have antimicrobial activity *(6, 10)*. However, testing many different product formulations for a variety of acidified foods to demonstrate a 5-log reduction in bacterial pathogen levels may be impractical. Even small changes in product formulation would require new microbiological tests to determine safe processing conditions. Variation in the concentration and efficacy of these antimicrobial compounds may occur due to growing conditions of the source plant material and processing procedures. Therefore, we focused our studies on ingredients, such as acetic acid and benzoic acid, that would be common to many acidified food products

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TABLE 1. *Bacterial strains used in this study*

Study strain ID	Strain name	Previous ID[a]	Source
B200	*E. coli* O157:H7	ATCC 43888	Laboratory
B201	*E. coli* O157:H7	SRCC 1675	Apple cider
B202	*E. coli* O157:H7	SRCC 1486	Salami outbreak
B203	*E. coli* O157:H7	SRCC 206	Ground beef
B204	*E. coli* O157:H7	SRCC 1941	Pork
B206	*S. enterica* Braenderup	SRCC 1093	10% salted yolk
B207	*S. enterica* Cerro	SRCC 400	Cheese powder
B208	*S. enterica* Enteritidis	SRCC 1434	Ice cream
B209	*S. enterica* Newport	SRCC 551	Broccoli with cheese
B210	*S. enterica* Typhimurium	SRCC 1846	Liquid egg
B0195	*L. monocytogenes*	SRCC 529	Pepperoni
B0196	*L. monocytogenes*	SRCC 1791	Yogurt
B0197	*L. monocytogenes*	SRCC 1506	Ice cream
B0198	*L. monocytogenes*	SRCC 1838	Cabbage
B0199	*L. monocytogenes*	SRCC 2075	Diced coleslaw

[a] SRCC, strains obtained from Silliker, Inc. (Chicago, IL); ATCC, American Type Culture Collection (Manassas, VA).

and for which the antimicrobial activity could be consistently determined. For these studies, we used a noninhibitory vegetable broth medium. Cucumber juice from pasteurized brined cucumbers has been used for this purpose in previous studies of the safety of acidified foods because this vegetable-based medium contains nutrients (sugars and amino acids) that may aid survival of bacterial pathogens. Cucumber juice also contains no known antimicrobial compounds and therefore can be used for a ''worst-case'' scenario for the survival of pathogens in challenge studies *(1, 2)*.

The objectives of this study were to determine a variety of safe processing conditions for acidified foods with pH values of 3.5 and 3.8, conditions representative of dressing products and other acidified foods. Because the survival of vegetative bacterial pathogens in acidified foods is enhanced as temperature decreases, we choose 10°C for these studies. Foods held at colder temperatures could be considered refrigerated foods, which are exempt from acidified food regulations and are beyond the scope of this work. The effect of the added acid would be more detrimental to microbial survival at higher temperatures, therefore 10°C represents a worst-case scenario. The majority of the work presented here was done with a cocktail of *E. coli* O157:H7 strains isolated from foods. Previous results have indicated that enterohemorrhagic *E. coli* strains are the most acid-resistant pathogens of concern in acidified foods *(1, 2)*. We report here on the conditions needed for a 5-log reduction of pathogenic *E. coli* in cucumber juice medium containing combinations of acetic and benzoic acids at pH values of 3.5 and 3.8. These results were also confirmed with strains of *S. enterica* and *L. monocytogenes*.

MATERIALS AND METHODS

Preparation of bacterial cells. Cocktails consisting of five strains each of the foodborne pathogens *E. coli* O157:H7, *S. enterica,* and *L. monocytogenes* (Table 1) were used in these experiments. Isolated colonies of each strain were inoculated separately into 5 ml of Luria-Bertania (LB) broth (Difco, BD, Sparks, MD) for *E. coli* and *Salmonella* or tryptic soy agar (TSA; Difco) for *L. monocytogenes*. Growth medium was supplemented with 1% glucose, and cultures were grown statically (to induce acid resistance) in triplicate in separate 15-ml screw-cap tubes and incubated at 37°C for 16 h. After growth, the cells were harvested by centrifugation (2,000 × *g,* 10°C; Sorvall RC-5B, SS-34 rotor) and then resuspended in 0.5 ml of sterile saline (0.85% NaCl). Cell cultures of the same species were combined using equal amounts (0.5 ml) of each strain. Approximately 2 ml of the concentrated cell suspension was inoculated into each jar of brined cucumbers (see below) to give an initial cell count of approximately 10^8 CFU/ml. At time intervals of 24 h or less (as indicated), a 1-ml sample of the inoculated brine was removed through a 15-mm septum in the jar lid *(1)* and diluted 10-fold into 50 mM 3-*N*-morpholinopropanesulfonic acid buffer (pH 7) to neutralize the pH. The buffered cell suspensions were subsequently diluted in 0.85% NaCl, and the culture population was determined by plating using a spiral plater (Spiral Biotech, Inc., Norwood, MA) and LB agar (LB containing 1.5% agar; Fisher Scientific, Fair Lawn, NJ). Petri plates were incubated for 24 to 48 h at 37°C, and the colonies were counted with an automated spiral plate counter (Q-Count, Spiral Biotech). All chemicals were obtained from Sigma-Aldrich (St. Louis, MO) unless otherwise specified.

Preparation of pickle brines. Size 2B (about 3.75 cm in diameter) cucumbers were received from a local source and used for the preparation of acidified pickles. After washing, approximately 420 g of cucumbers was packed with 420 ml of brine in 0.85-liter glass jars. The brine consisted of an organic acid (acetic or citric acid, as indicated), 4% NaCl, and 0.2% $CaCl_2$ per jar, to equilibrate at the desired acid concentration (2% NaCl, 0.1% $CaCl_2$). To achieve the desired pH (after equilibration), equal weights of cucumbers and brine were blended and titrated with 1 N NaOH to determine volumes of NaOH needed per jar to achieve the desired pH of 3.5 or 3.8. Jars were sealed and pasteurized in a water bath to an internal temperature of 75°C for 15 min. The internal temperature was determined by inserting a metal thermometer through the septum in the jar lid into the cold center cucumber in the jar. The jars were cooled to ambient temperature (23 to 25°C) and allowed to rest for at least 10 days to ensure equilibration of water-soluble elements (i.e., salts, acids, and sugars) between the cucumbers and the acid solution. Brine samples (1 ml) were withdrawn from jars through the lid septum for

biochemical analysis by high-performance liquid chromatography (HPLC) (as described below) to verify the acetic and benzoic acid concentrations and pH. The jars were then incubated for 2 days at 10°C to ensure temperature equilibration before inoculation with pathogens.

Biochemical analysis. The pH of brine samples was determined with an IQ240 pH meter (IQ Scientific Instruments, San Diego, CA). HPLC analysis was used to determine organic acid concentrations using a modification of the method of McFeeters and Barish *(14)*. Organic acid concentrations were measured with a Thermo Separation Product HPLC system (ThermoQuest, Inc., San Jose, CA) consisting of a P2000 pump, an SCM100 solvent degasser, an AS3000 autosampler, and a UV6000 diode array detector. An HPX-87H column (300 by 7.8 mm; Bio-Rad Laboratories, Hercules, CA) was used to resolve acetic and benzoic acids. The operating conditions of the system were a column temperature of 37°C and 0.03 N H_2SO_4 eluent at a flow rate of 0.6 ml/min. The UV6000 detector was set to 210 nm at a rate of 1 Hz for data collection. ChromQuest version 4.1 chromatography software was used to control the system and analyze the data, utilizing the peak heights for quantitative integration.

Modeling and statistical analysis. All experiments were carried out with three or more independent replicates. A single linear mixed effects model was used to analyze the 390 observations from 64 independent replicate runs of 15 different treatment conditions of acid type, acid concentration, pH, and organism. The model involved 15 intercepts and 15 slopes for these combinations. Because repeated measures were made over time for each replicate, a correlation structure was imposed by adding random coefficients (corresponding to three or more replicate runs) to the slopes. The model may be written as

$$Y_{ij}(t_k) = \beta_0{}^{ij} + (\beta_1{}^{ij} + U_{ij})t_k + e_{ijk}$$

where $Y_{ij}(t_k)$ is the log CFU from replicate j of experimental condition i at time t_k and $\beta_0{}^{ij}$ and $\beta_1{}^{ij}$ represent the regression parameters (here the superscripts represent a notational index and not an exponent). Random coefficients are incorporated through the U_{ij} terms, which are assumed to be a random sample from the normal distribution with mean 0 and variance component $\sigma^2{}_U$. The 5-log reduction times (days) were estimated using the reciprocal of the generalized least squares estimate of the slopes obtained using PROC MIXED ($5D = -5/\hat{\beta}_i$). Because these values are nonlinear functions of parameter estimates, approximate standard errors for the estimated 5-log reduction times were computed using the delta method, based on a first-order Taylor's approximation of the standard error of $1/\hat{\beta}$ as

$$\text{SE (5-log reduction time)} = 5\text{SE}\left(\hat{\beta}\right)/\left(\hat{\beta}^2\right)$$

Pairwise tests of equality of the mean 5-log reduction times were conducted at a significance level of 0.01.

RESULTS AND DISCUSSION

A critical factor for the safety of acidified foods is the destruction of vegetative cells of microorganisms of public health significance, as defined under 21 CFR 114 *(20)* and FSMA *(19)*. To assure safety, producers of acid and acidified vegetable products that are not pasteurized can achieve 5-log reduction in bacterial pathogens by holding acidic products under a defined time-temperature regimen. Although heat processing can achieve a 5-log reduction, heating some products, such as dressings, mayonnaise products, and pickled peppers, can result in undesirable sensory properties. Previous data indicate that temperature is a critical factor influencing the freshness of acidified vegetable products in acid and that colder temperatures lead to longer shelf life *(1)*. Therefore, we chose 10°C for these studies (above refrigeration) as a worst-case scenario. Similarly, we induced acid resistance for cocktails of bacterial strains by static growth in the presence of a fermentable sugar (glucose). The acid produced during growth can serve to induce acid resistance *(4)*, and the survival of acid-injured cells can be encouraged by plating survivors on a nonselective medium (LB agar). Anaerobic conditions promote survival of pathogenic *E. coli* strains in acid solutions *(12)*; therefore, our studies were carried out in pasteurized (anaerobic) sealed jars of brined cucumbers. The cucumber brine medium in the pasteurized jars contained sugars and amino acids common to many acidified foods, which may enhance survival of *E. coli* through amino acid decarboxylation *(7)*.

Acetic acid concentrations for this study were chosen based on suggestions from manufacturers of dressings, mayonnaise, pickled peppers, and other acidified food products. Many of these products contain both an oil or lipid phase and an aqueous phase. For dressings, the aqueous phase is typically around 55% (vol/vol), and mayonnaise typically has a 10 to 12% aqueous phase. The acidity in these products, concentrated in the aqueous phase, can range from 1.5 to 2.5% acetic acid or higher, with pH values of 3.3 to 3.8 or greater. Vegetative bacterial pathogens may be present in the aqueous phase of these products (which are not heat treated). Other models for inactivation of *E. coli* in foods have been developed *(5, 15)*, but published data to support process filings on the holding times and temperatures needed for a 5-log reduction in vegetative pathogen numbers in acidified foods are lacking.

The inocula used in this study, i.e., cocktails of *E. coli* O157:H7, *Salmonella*, and *L. monocytogenes*, were previously used for similar studies *(1, 3)* cited in U.S. Food and Drug Administration (FDA) process filings for commercial production of acidified foods. Related serotypes, including the *E. coli* O104:H4 European outbreak strain, have acid resistance similar to that of the strains used in the present study *(16)*. For all acid conditions tested, *E. coli* O157:H7 strains were more acid resistant than *S. enterica* and *L. monocytogenes* cocktails (Table 2), in agreement with previously published data *(1)*. The times needed for a 5-log reduction of *S. enterica* or *L. monocytogenes* were two to three times shorter than those for needed for *E. coli* O157:H7 strains at pH 3.8 and 3.5 for selected acid conditions (Fig. 1). At pH 3.5, the *L. monocytogenes* strains were significantly more sensitive to acid than were the *S. enterica* strains ($P > 0.01$). However, there was no significant difference in the survival of *L. monocytogenes* and *S. enterica* strains in the pH 3.8 treatment (2.5% acetic acid plus 0.1% benzoic acid) ($P > 0.01$). Overall, reducing the pH from 3.8 to 3.5 reduced the 5-log reduction time. A 2.8-fold reduction in survival was observed for acid solutions containing 2.5% acetic acid when initial pH was

TABLE 2. *Acid solutions and 5-log reduction times of inoculated pathogens in brined cucumbers*

Species[a]	Acid concn (%)[b]		pH	5-log reduction time (days)		R^2[c]	No. of replicates
	Acetic acid	Benzoic acid		Mean	SE		
E. coli	2.5	0	3.5	4.0	0.23	0.88	12
	2	0	3.5	11.7	0.43	0.90	11
	2	0.1	3.5	1.5	0.05	0.99	3
	1.5	0.1	3.5	3.6	0.43	0.79	6
	0[d]	0.1	3.5	14.5	0.97	0.88	3
	2.5	0	3.8	11.3	0.75	0.90	3
	2.5	0.1	3.8	3.6	0.14	0.99	3
	2	0.1	3.8	10.2	0.51	0.95	3
	1.5	0.1	3.8	13.5	1.03	0.88	3
Salmonella	2.5	0	3.5	1.5	0.11	0.91	3
	2.5	0.1	3.8	1.6	0.08	0.97	3
	1.5	0.1	3.5	0.6	0.03	0.97	3
L. monocytogenes	2.5	0	3.5	0.6	0.03	0.95	3
	2.5	0.1	3.8	1.5	0.24	0.79	3
	1.5	0.1	3.5	0.3	0.01	0.98	3

[a] Each species cocktail contained five strains.
[b] All acid concentrations were ±0.1% of the indicated target concentrations.
[c] Value for the linear regression used to calculate the 5-log reduction time.
[d] This acid solution contained 0.5% citric acid buffer.

reduced from 3.8 to 3.5; 3.7- and 6.8-fold reductions were observed for 1.5% acetic acid plus 0.1% benzoate and 2% acetic acid plus 0.1% benzoate, respectively, for the same pH reduction (Table 2).

The use of preservatives is usually limited to inhibiting spoilage organisms in acidified foods. The acidified foods regulation 21 CFR 114 *(20)* states ''Permitted preservatives may be used to inhibit reproduction of microorganisms of non-health significance (in lieu of thermal processing).''

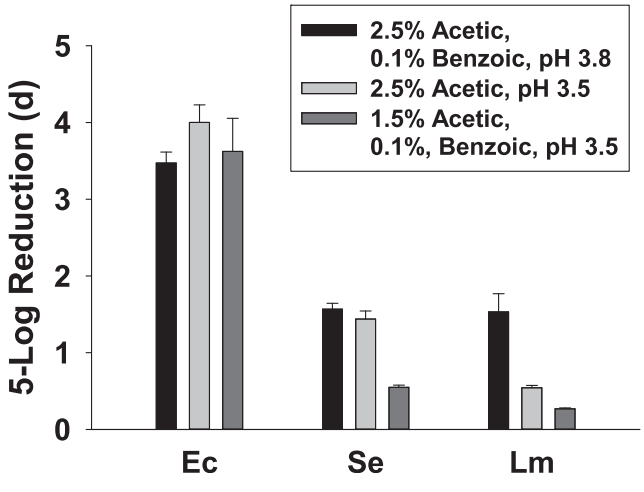

FIGURE 1. *Survival of* E. coli *O157:H7 (Ec),* S. enterica *(Se), and* L. monocytogenes *(Lm) in selected acid solutions. The 5-log reduction times (days) are shown for treatments with 2.5% acetic acid plus 0.1% benzoic acid, pH 3.8 (solid bars); 2.5% acetic acid, pH 3.5 (light shaded bars); and 1.5% acetic acid plus 0.1% benzoic acid, pH 3.5 (dark shaded bars). The error bars indicate the standard error of the estimate of time required for a 5-log reduction.*

This regulation does not explicitly state that preservatives cannot be used for assuring safety, although the regulation may be interpreted in that way. Currently, processors must list preservatives used and their concentration on forms when filing their processes with the FDA. However, results of several studies have indicated that preservative acids, including benzoate, sorbate, and sulfite, contributed to killing pathogenic bacteria in foods *(8, 9, 11, 18).* A comparative study of the relative concentrations of organic acids needed for a 5-log reduction of *E. coli* O157:H7 at pH 3.2 revealed that benzoic and sorbic acids were significantly more effective than common food acids (citric, malic, lactic, and acetic) for killing bacterial pathogens, based on equimolar protonated acid concentrations *(13).* Those authors also found that citric acid functioned primarily as a buffer and did not contribute directly (other than the effect of pH) to acid killing of *E. coli* O157:H7. We found that a 5-log reduction of *E. coli* O157:H7 occurred within 14.5 days at pH 3.5 in the presence of benzoic acid when citric acid was used as a buffer (Table 2). For acidified foods containing benzoate at pH 3.5 or lower, a 5-log reduction in bacterial pathogens could therefore occur in approximately 2 weeks (14.5 days) based on the benzoic acid concentration alone. In mixed acid solutions, the effect of the presence or absence of benzoate with two different acetic acid solutions is shown in Figure 2. The addition of benzoic acid decreased the 5-log reduction times threefold for 2% acetic acid at pH 3.5 and more than sevenfold for 2.5% acetic acid at pH 3.8 (Fig. 2). The 5-log reduction times for additional acetic and mixed acid (acetic and benzoic acids) conditions representative of acidified foods are shown in Table 2.

With the advent of the FSMA, critical controls for acidified foods may include a 5-log reduction in vegetative

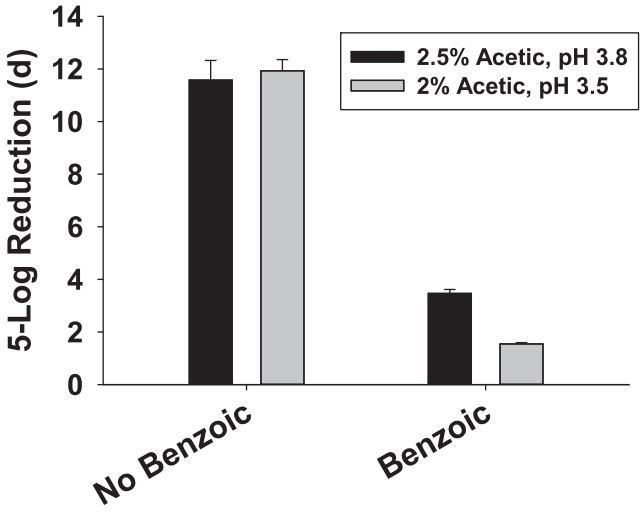

FIGURE 2. *Effect of benzoic acid on the survival of* E. coli *O157:H7 at pH 3.5. The 5-log reduction times (days) are shown for treatments with 2.5% acetic acid, pH 3.8 (solid bars) and 2% acetic acid, pH 3.5 (shaded bars) in the presence or absence of 0.1% benzoic acid. The error bars indicate the standard error of the estimate of time required for a 5-log reduction.*

bacterial pathogens such as *E. coli* O157:H7, *S. enterica,* and *L. monocytogenes*. The experiments in the present study were conducted in a noninhibitory vegetable broth medium at 10°C with acid-adapted pathogen cells, representing a worst-case scenario for a variety of acidified food products. The *E. coli* O157:H7 strains selected had acid resistance typical of a variety of enterohemorrhagic bacterial strains from foods, although the strains used were not necessarily from outbreaks associated with produce *(17)*. The 5-log reduction times for pathogenic bacteria in acidified foods containing acetic and benzoic acids at pH 3.5 and 3.8 revealed that acid killing was dependent on the concentration and type of acid, and the preservative benzoic acid significantly reduced the time needed to achieve a 5-log reduction. We also estimated the standard error of the mean 5-log reduction times. Previously, five times the standard error estimate was added to the 5-log reduction times as an arbitrary safety factor for thermal processes *(2)* and cold fill-hold data *(1)*. For recommended hold times for acidified foods based on the current study, a competent process authority should be consulted to determine whether any safety factor in addition to the measured 5-log reduction times is needed.

ACKNOWLEDGMENTS

This study was supported in part by research grants from Marcatus QED (Toronto, Ontario, Canada), Cargill, Inc. (Minnetonka, MN), Edco Food Products, Inc. (Hobart, WI), U.S. Department of Agriculture (USDA) National Integrated Food Safety Initiative 2011-51110-31019, Pickle Packers International, Inc. (Washington, DC), and USDA Cooperative State Research, Education, and Extension Service project 2006-01240. We thank Ms. Sandra Parker for excellent editorial assistance.

REFERENCES

1. Breidt, F., Jr., J. Hayes, and R. F. McFeeters. 2007. Determination of 5-log reduction times for food pathogens in acidified cucumbers during storage at 10 and 25°C. *J. Food Prot.* 70:2638–2641.
2. Breidt, F., Jr., J. S. Hayes, J. A. Osborne, and R. F. McFeeters. 2005. Determination of 5-log pathogen reduction times for heat-processed, acidified vegetable brines. *J. Food Prot.* 68:305–310.
3. Breidt, F., K. P. Sandeep, and F. Arritt. 2010. Use of linear models for thermal processing of acidified foods. *Food Prot. Trends* 30:268–272.
4. Buchanan, R. L., and S. G. Edelson. 1999. pH-dependent stationary phase acid resistance response of enterohemorrhagic *Escherichia coli* in the presence of various acidulants. *J. Food Prot.* 62:211–218.
5. Champan, B., K. J. Scurrah, and T. Ross. 2010. Contemporary formulation and distribution practices for cold-filled acid products: Australian industry survey and modeling of published pathogen inactivation data. *J. Food Prot.* 73:895–906.
6. Davidson, P. M., and T. M. Taylor. 2007. Chemical preservatives and naturally antimicrobial compounds, p. 713–745. *In* M. P. Doyle and L. R. Beuchat (ed.), Food microbiology: fundamentals and frontiers, 3rd ed. ASM Press, Washington, DC.
7. Foster, J. W. 2004. *Escherichia coli* acid resistance: tales of an amateur acidophile. *Nat. Rev. Microbiol.* 2:898–907.
8. Hwang, C.-A., and L. R. Beuchat. 1995. Efficacy of lactic acid/sodium benzoate wash solution in reducing bacterial contamination of raw chicken. *Int. J. Food Microbiol.* 27:91–98.
9. Ingham, S. C., and N. P. Schoeller. 2002. Acceptability of a multi-step intervention system to improve apple cider safety. *Food Res. Int.* 35:611–618.
10. Juneja, V. K., H. P. Dwivedi, and X. Yan. 2012. Novel natural food antimicrobials. *Annu. Rev. Food Sci. Technol.* 3:381–403.
11. Juneja, V. K., S. T. Martin, and G. M. Sapers. 1998. Control of *Listeria monocytogenes* in vacuum-packaged pre-peeled potatoes. *J. Food Sci.* 63:1–4.
12. Kreske, A. C., K. Bjornsdottir, F. Breidt, Jr., and H. Hassan. 2008. Effects of pH, dissolved oxygen, and ionic strength on the survival of *Escherichia coli* O157:H7 in organic acid solutions. *J. Food Prot.* 71: 2404–2409.
13. Lu, H. J., F. Breidt, Jr., I. M. Pérez-Díaz, and J. A. Osborne. 2011. The antimicrobial effects of weak acids on the survival of *Escherichia coli* O157:H7 under anaerobic conditions. *J. Food Prot.* 6:893–898.
14. McFeeters, R. F., and A. O. Barish. 2003. Sulfite analysis of fruits and vegetables by high-performance liquid chromatography (HPLC) with ultraviolet spectrophotometric detection. *J. Agric. Food Chem.* 51:1513–1517.
15. McQuestin, O. J., C. T. Shadbolt, and T. Ross. 2009. Quantification of the relative effects of temperature, pH, and water activity on inactivation of *Escherichia coli* in fermented meat by meta-analysis. *Appl. Environ. Microbiol.* 75:6963–6972.
16. Oh, D.-H., F. Breidt, Jr., and P. M. Fratamico. Unpublished data.
17. Oh, D.-H., Y. Pan, E. Berry, M. Cooley, R. Mandrell, and F. Breidt, Jr. 2009. *Escherichia coli* O157:H7 strains isolated from environmental sources differ significantly in acetic acid resistance compared with human outbreak strains. *J. Food Prot.* 72:503–509.
18. Samellis, J., J. N. Sofos, M. L. Kain, J. A. Scanga, K. E. Belk, and G. C. Smith. 2001. Organic acids and their salts as dipping solutions to control *Listeria monocytogenes* inoculated following processing of sliced pork bologna stored at 4°C in vacuum packages. *J. Food Prot.* 64:1722–1729.
19. U.S. Food and Drug Administration. 2011. Food Safety Modernization Act. Available at: http://www.fda.gov/Food/FoodSafety/FSMA/ucm247548.htm. Accessed 7 March 2013.
20. U.S. Food and Drug Administration. 2012. Food for human consumption. Acidified foods. 21 CFR 114. U.S. Food and Drug Administration, Washington, DC.