

Evaluation of Small-Scale Hot-Water Postpackaging Pasteurization Treatments for Destruction of *Listeria monocytogenes* on Ready-to-Eat Beef Snack Sticks and Natural-Casing Wieners

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MS 05-62: Received 15 February 2005/Accepted 16 May 2005

ABSTRACT

This study was conducted to evaluate small-scale hot-water postpackaging pasteurization (PPP) as a postlethality (post-cooking) treatment for *Listeria monocytogenes* on ready-to-eat beef snack sticks and natural-casing wieners. Using a commercially available plastic packaging film specifically designed for PPP applications and 2.8 liters of boiling water (100°C) in a sauce pan on a hot plate, an average reduction in *L. monocytogenes* numbers of ≥ 2 log units was obtained using heating times of 1.0 min for individually packaged beef snack sticks (three brands) and 4.0 min for packages of four sticks (two brands) and seven sticks (three brands). Average product surface temperatures, measured as soon as possible after PPP and opening the package, were 47 to 51.5, 58 to 61.5, and 58.5 to 61°C for the beef snack sticks packages of one, four, and seven sticks per package, respectively. A treatment of 7.0 min for packages of four natural-casing wieners (three brands) achieved *L. monocytogenes* reductions of ≥ 1.0 log unit and average product surface temperature of 60.5 to 63.5°C. Cooked-out fat and moisture resulting from tested treatments ranged from 0.2 to 1.1% by weight for beef snack sticks and from 0.4 to 1.2% by weight for natural-casing wieners. For natural-casing wieners, PPP had no detrimental effect on overall product desirability to consumers; results suggested that PPP may significantly enhance appearance of this product. However, for beef snack sticks the cooking out of fat and moisture during PPP had a significant negative effect on consumer opinions of product appearance.

On 6 June 2003, the U.S. Department of Agriculture (USDA) published an interim final rule addressing the control of *Listeria monocytogenes* on ready-to-eat (RTE) meat and poultry products (9). This rule went into effect on 6 October 2003 and was intended to encourage processors of RTE products to take one or more specific steps to ensure the absence of *L. monocytogenes* on their products. Possible steps range from using focused sanitation procedures to adding ingredients or using processing treatments designed to kill *L. monocytogenes* or inhibit its growth. Under the regulations, the processor is also required to perform testing for *L. monocytogenes* or other *Listeria* spp. on food contact surfaces in the area of the plant in which RTE products are handled after cooking. The amount of testing is related to the types of RTE products made, the product ingredients, and how the products are processed and handled. In particular, the rule requires processors of RTE meat and poultry products to adopt one of three designated alternatives for controlling *L. monocytogenes* on their products. The alternatives involve various levels of control and microbiological testing. Under alternative 1, the processor uses a postlethality treatment that reduces or eliminates *L. monocytogenes* and an antimicrobial agent or process that suppresses or limits *L. monocytogenes* growth throughout product shelf

life. Under alternative 2, the processor uses either a postlethality treatment that reduces or eliminates *L. monocytogenes* or an antimicrobial agent or process that suppresses or limits *L. monocytogenes* growth throughout product shelf life. Under alternative 3, only sanitation measures are relied upon to control *L. monocytogenes*.

The objective of this work was to evaluate small-scale hot-water postpackaging pasteurization (PPP) for use as a postlethality treatment by small and very small processors under alternatives 1 or 2 of the USDA regulations. This postlethality treatment was studied for use with beef snack sticks and natural-casing wieners, two products commonly made by small and very small processors. In the processing of beef snack sticks, the addition of salt and the processes of cooking and smoking are effective antimicrobial interventions that make the finished product unsuitable for *L. monocytogenes* growth (4). Some processors also acidify beef snack sticks, either through fermentation or through the addition of an acidulant. Either means of acidification would provide further inhibition of *L. monocytogenes* growth. For beef snack sticks, the adoption of hot-water PPP as an effective postlethality treatment could allow processors to operate under alternative 1. The second product studied, natural-casing wieners (finely ground cured sausage contained inside an intestine casing, often used to make a “hot dog” sandwich), has traditionally been made without the addition of any ingredients that effectively in-

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hibit *L. monocytogenes* growth. Hot-water PPP of this product would allow processors to operate under alternative 2. However, many processors have begun adding an antimicrobial agent such as sodium lactate to their natural-casing wiener recipes. For these processors, the use of hot-water PPP would allow operation under alternative 1.

There are a large number of variables involved in hot-water PPP, including packaging film composition and thickness, mass of product, volume of water, proportions of water and treated product, water temperature, and treatment time. To simplify potential techniques that small and very small processors might adopt, we focused our efforts by using a single type of packaging film, typical masses and packaging configurations of products, a single amount of hot water, and a single initial water temperature. We varied the treatment time, keeping in mind that processors would prefer shorter treatments for reasons of economics and efficiency.

We also paid considerable attention to whether effective hot-water PPP treatments affected the sensory properties of beef snack sticks and natural-casing wieners. Because these products would be subjected to a thermal process in a confining package, cook-out of product components (almost entirely fat) was determined to be a potential drawback of this procedure. Therefore, we measured the amount of material cooked out and conducted consumer sensory panels of treated and untreated products.

MATERIALS AND METHODS

Meat products. Three brands each of beef snack sticks and natural-casing wieners were purchased at local grocery stores and transported within 30 min to the laboratory. Samples were refrigerated ($5 \pm 1^\circ\text{C}$) until use. A sample from a randomly chosen lot of each product was analyzed in the laboratory for water activity using a Decagon water activity meter (AquaLab Series 3 TE, Pullman, Wash.) and for pH (sample homogenized in distilled water) using an Accumet AB15 pH meter (Fisher Scientific, Itasca, Ill.) with a probe. Samples also were sent to a commercial laboratory to be analyzed for moisture, salt, and fat percentages using forced-air-oven determination of moisture (AOAC method 950.46Bb), potentiometric method for salt (AOAC method 980.25), and Soxhlet method for fat (AOAC method 960.39) (2). The moisture:protein ratio and percentage of water-phase salt were calculated from these analytical results. Representative products were measured to determine average length and diameter. Physical and chemical characteristics of the products are given in Table 1.

Bacterial cultures and preparation of inoculum. The *L. monocytogenes* strains used in this study were obtained from the laboratory of Dr. Eric Johnson (Food Research Institute, University of Wisconsin–Madison). Strain Scott A was a clinical isolate, strains LM 101 and LM 108 were isolated from hard salami, strain LM 310 was isolated from goat cheese, and strain V7 was isolated from raw milk. Stock cultures were maintained at -20°C in brain heart infusion broth (BHIB; Difco, Becton Dickinson, Sparks, Md.) with 10% (wt/vol) glycerol (Fisher) added. Working cultures maintained at 4°C on brain heart infusion agar (BHIA; Difco, Becton Dickinson) were prepared monthly from frozen stock cultures. To obtain a working culture, a strain was cultured twice successively at 35°C for 18 to 24 h in BHIB, streaked onto a BHIA plate, incubated at 35°C for 18 to 24 h, examined for purity, and then stored at 4°C . Inoculation cultures were prepared for each

TABLE 1. Physical and chemical characteristics of commercial beef snack sticks and natural-casing wieners prior to hot-water postpackaging pasteurization

Product (processor)	Mean preheat length (cm)	Mean preheat diameter (cm)	pH	Water activity	Moisture (%)	Protein (%)	Moisture: protein	Water phase salt (%)	Fat (%)	Typical sample weight (g) ^a		
										1/package	4/package	7/package
Beef snack sticks												
A	20	1.3	4.5	0.91	36.0	20.6	1.7	9.6	35.6	4.5	18.0	31.0
B	12.6	1.6	5.0	0.86	29.1	17.2	1.7	8.8	46.4	6.0	23.0	40.0
C	15.1	1.4	5.0	0.89	34.6	20.0	1.7	10.1	36.6	4.5	18.0	31.0
Natural-casing wieners												
A	12	2.1	5.9	0.95	53.6	12.3	4.3	3.6	27.1		44.0	
B	13.2	2.4	6.1	0.97	52.3	12.8	4.1	3.8	31.6		43.0	
C	13.2	2.5	6.3	0.97	58.5	12.3	4.7	3.1	25.7		41.0	

^a Sample consisted of one 2.5-cm longitudinal segment from the middle of each piece of product and was obtained by making two cuts in the plane of the product diameter.

strain by transferring a loopful of growth from the working culture plate to 9 ml of BHIB and incubating at 35°C for 20 to 24 h. To prepare the five-strain inoculum cocktail, the BHIB cultures were combined into a 50-ml sterile plastic centrifuge tube and centrifuged for 10 min at $5,000 \times g$. The supernatant in the tube was decanted, and the pellet was resuspended to the original volume in Butterfield's phosphate diluent (BPD; Nelson Jameson, Marshfield, Wis.) to make an inoculum cocktail. The inoculum cocktail was serially diluted in BPD and plated to determine cell concentration (ca. 8 log CFU/ml).

Inoculation of meat products. Two different methods were used to inoculate products. Natural-casing wieners and beef snack sticks to be packaged four or seven-per-package were inoculated by placing 110 ml of a 1:10 dilution (in BPD) of inoculum cocktail in a large glass petri dish and rolling an individual product piece through the inoculum for two complete revolutions. This procedure was used to ensure that in situations where product piece-to-piece contact would occur all product surfaces were inoculated, thus maximizing the opportunity for protection of cells via piece-to-piece contact. Individually packaged beef sticks do not experience piece-to-piece contact within the package, so each beef snack stick to be packaged individually was inoculated by placing 25 μ l of the 1:10 dilution (BPD) of inoculum cocktail on each of five sites along the beef snack stick. The inoculum was then spread across the beef snack stick surface using a sterile plastic spreader. After inoculation, products were allowed to dry at room temperature (20 to 22°C) for 90 min in a biosafety level 2 hood before packaging. The mean starting inoculum concentrations in initial trials were 4.2 log CFU per sample for beef snack sticks (range, 3.1 to 5.3; standard deviation, 0.7) and 5.9 log CFU per sample for natural-casing wieners (range, 5.7 to 6.3; standard deviation = 0.2). In confirmatory trials, mean starting inoculum concentrations were 3.9 log CFU per sample for beef sticks (range, 2.7 to 5.5; standard deviation = 0.7) and 5.7 log CFU per sample for natural-casing wieners (range, 4.7 to 6.1; standard deviation = 0.4).

Packaging of meat products. A commercial plastic film designed for use in hot-water or steam PPP (product SPP94, material no. CPS302976, Curwood, Inc., New London, Wis.) was made into bags that were custom sized for products using scissors and a heat sealer on a commercial vacuum packaging machine. Dry inoculated products were placed in appropriate bags and vacuum packaged (at approximately 1 atm). Trials were done with one, four, and seven beef snack sticks per package and with four natural-casing wieners per package.

Hot-water PPP treatments. Prior to pasteurization, 2.8 liters of tap water was placed in an aluminum sauce pan 21 cm in diameter and 12 cm tall. A plastic test tube rack was submerged in the pan using a lead donut-shaped weight. This rack was used to hold the packaged product off of the pan bottom during heating and thus allow maximum water contact with the product. Either of two hot plates was used to heat the water to boiling for the pasteurization trials. The hot plates had maximum power of 1,113 and 1,118 watts. A single package of inoculated product was placed between levels of the test tube rack in the boiling water. Although the water stopped boiling when packaged product was first added, it resumed boiling within a short time (<30 s for the largest packages). After the prescribed time had elapsed, the package was immediately removed from the water and placed in ice-water slush. The next sample was not processed until after the water had again boiled. Additional water was added to the pan as needed to maintain water volume. An initial trial consisted of

three packages of product, all inoculated with an inoculum cocktail from the same broth cultures, for each of several treatment times. A confirmatory trial consisted of one package of product per treatment. Untreated control packages were handled identically to treated packages other than during submersion in the boiling water. Initial trials for packages of one, four, and seven beef snack sticks were conducted for 0.5, 1.0, and 1.5; 3.0, 4.0, and 5.0; and 3.0, 4.0, 5.0, and 6.0 min, respectively. Initial trials with the packages of four natural-casing wieners were conducted for 5.0, 6.0, and 7.0 min. Confirmatory trials were conducted with treatments of 1.0 and 4.0 min for packages of one and of four or seven beef snack sticks, respectively, and for 7.0 min for packages of four natural-casing wieners.

Enumeration of inoculum bacteria. Each chilled package of product was removed from the ice-water slush, and the package surface was patted dry with a paper towel and then sprayed with 70% ethanol. Following a 20-min drying period, each package was aseptically opened. The ends of each beef snack stick or natural-casing wiener in the package were removed, leaving 2.5-cm middle pieces that were used as the sample (typical sample weights are shown in Table 1). The samples and the package material minus remaining product were transferred to the stomacher filter bag. A standard 198 ml of BPD was added to the stomacher bag, and the contents were manually massaged for 1 min and then manually shaken for 1 min. This procedure ensured thorough contact of BPD with product and package interior surfaces. From the initial sample dilution (arbitrarily denoted as 10^{-2}), further dilutions were made as appropriate using BPD. For initial trials, no effort was made to allow repair and subsequent enumeration of injured cells. From the initial dilution, 1.0 ml was distributed for spread plating among three plates (0.3, 0.3, and 0.4 ml) of *Listeria* selective agar (LSA; Oxoid, Ogdensburg, N.Y.) with *Listeria* selective supplements (Oxford formulation; Oxoid). From the original dilution and each subsequent dilution, 0.1 ml was spread onto one LSA plate per dilution. Plates were incubated at 35°C for 48 h according to the manufacturer's instructions and then examined for typical *L. monocytogenes* colonies (small-to-medium brown-to-black colonies surrounded by a black precipitate zone), which were counted. For each product tested in each trial, one presumptive *L. monocytogenes* colony was selected for confirmation testing. The colony was transferred to BHIA for culture and then tested for the Gram stain reaction, oxidase activity, and biochemical characteristics (API *Listeria* kit, bioMérieux, Hazelwood, Mo.), and cellular morphology was examined. Throughout the study, all presumptive isolates were confirmed as *L. monocytogenes*. For confirmatory trials, dilutions were spread plated on BHIA and incubated for 1 h at 35°C (unpublished method) to allow repair of injured cells. Following this incubation period, each BHIA plate was then overlaid with about 10 ml of tempered (46°C) LSA plus supplements and then incubated for 48 h at 35°C. For each trial, the difference in log CFU between treated and untreated product was considered the treatment lethality. When no colonies resulted from the least dilute plates (arbitrary 10^{-2} dilution), a value of nine colonies on a hypothetical 10^{-1} dilution plate (effectively one colony less than the detection limit) was assigned. The log of 90 CFU, rounded to 1.9, was then used in calculating treatment lethality.

Statistical analysis of microbiological data. The two-sample *t* test (release 14, Minitab, State College, Pa.) with a 5% significance level was used to make the following comparisons for log CFU reduction in initial trials: (i) processor versus processor for a given combination of product, number of pieces per package, and processing time; (ii) heating time versus heating time for a

given combination of product, processor, and number of pieces per package; and (iii) heating time versus heating time for a given combination of product and number of pieces per package (data for three processors combined). For confirmatory trials, the same analysis was done to compare processor versus processor for a given combination of product, number of pieces per package, and heating time.

Determination of product surface temperature. Using conditions otherwise identical to those in the initial and confirmatory trials, additional experiments were done with uninoculated products to determine product surface temperature at various times during hot-water PPP. At the designated time, the package of product was rapidly removed from the hot water and opened with a knife, and three product surface temperatures were rapidly measured at the midpiece (same location from which samples were obtained for microbiological analysis) using an infrared noncontact thermometer (Mini Temp FS, Raytek, Santa Cruz, Calif.). Particularly for multipiece packages, the product size and shape and piece-to-piece contact resulted in a range of surface temperatures. Therefore, the temperature was determined at three different midpiece locations, and the three temperatures were averaged for that package as follows. For individually packaged beef snack sticks, the temperature was measured at three places evenly around the beef snack stick circumference at the midpoint. For packages of four beef snack sticks and four natural-casing wieners, the temperature was measured on the upper surface midpiece for pieces 1 and 2 (counting from left to right), and piece 2 was rotated to obtain the surface temperature at the midpiece where piece 2 had contacted piece 3. For packages of seven beef snack sticks, surface temperatures were obtained on the upper surface midpiece for pieces 1 and 3, and piece 3 was rotated to obtain the surface temperature at the midpiece where piece 3 had contacted piece 4. For each combination of product, processor, number of pieces per package, and heating time, duplicate three-temperature averages were obtained and a mean of the duplicates was calculated.

Prediction of process lethality using surface temperature data. The lethality of the hot-water PPP process was estimated by inputting average surface temperature and time data into the American Meat Institute Foundation process lethality spread sheet (1). Along with the time and temperature data, a reference temperature of 62.8°C (145°F) and a z -value of 7.9°C (14.2°F), as determined by Muriana et al. (6) for *L. monocytogenes* in purge from smoked ham, were entered into the spreadsheet. These values were chosen because smoked ham was closest in composition to the beef snack sticks and natural-casing wieners of the three products studied by Muriana et al. (6). The F_0 values determined using the spreadsheet were divided by the previously determined $D_{62.8}$ -value of 67.6 s (6) for *L. monocytogenes* in purge from smoked ham to obtain a predicted log reduction.

Sensory quality of treated products. The percentage of product weight lost as cooked-out fat and water was determined for single trials of uninoculated beef snack sticks heated for 3.0 and 4.0 min (four beef snack sticks per package) and for 4.0 and 5.0 min (seven beef snack sticks per package) and for single trials of uninoculated natural-casing wieners (four per package) heated for 5.0, 6.0, and 7.0 min. Unscreened untrained panelists (178 to 192 individuals) also evaluated treated (4 min for package of seven beef snack sticks and 7 min for packages of four natural-casing wieners) and untreated beef snack sticks or natural-casing wieners. The beef snack sticks were presented to panelists with cooked-out fat and water removed. The treated and control natural-casing

wieners were separately warmed after being removed from the packaging by being placed under boiling water for 4.0 min and then held warm in an insulated container prior to serving. Panelists were self-selected from consumers entering a campus facility serving lunch and ice cream. Samples were presented in random order in cups coded with a three-digit random number. The panelist ballot contained two structured seven-point hedonic scales with possible scores ranging from 1 (dislike very much) to 7 (like very much). Mean scores for treated and untreated products were calculated, and an analysis of variance appropriate for a randomized complete block design (8) was conducted. Follow-up sensory evaluations were conducted with beef snack sticks and natural-casing wieners from processor B (rated highest by panelists in the initial sensory surveys). For the follow-up sensory evaluations, the packages of treated and untreated products were opened, and the packaging film was lifted from the product surface so that cooked-out fat and water were visible. The film surface was then loosely placed back over the product surface, and the bag was resealed by stapling. Panelists (198 individuals) at the same location were asked to evaluate the product appearance of control and treated products on a ballot with two structured seven-point hedonic scales with possible scores ranging from 1 (extremely unappealing) to 7 (extremely appealing). Mean scores for treated and untreated products were calculated, and an analysis of variance appropriate for a randomized complete block design (8) was conducted.

RESULTS AND DISCUSSION

In initial trials with beef snack sticks packaged one or four pieces per package, the average *L. monocytogenes* lethality increased with heating time (Table 2), with some significant differences between heating treatments. Specifically, increasing the heating time from 0.5 to 1.5 min for individually packaged beef snack sticks and from 3.0 to 4.0 or 5.0 min for packages of four sticks resulted in significantly greater lethality. Lethality differences between treatment times for beef snack sticks produced by a single processor were seldom significant for individually packaged and four-per-package product. Significant differences in lethality for beef snack sticks from different processors were also rare. For packages of seven beef snack sticks, a significant increase in lethality was achieved when the heating time was increased from 3.0 to 4.0 min (Table 2). Further increases in heating time did not result in a corresponding increase in lethality because of the plating detection limit being reached for PPP treatments of ≥ 4.0 min. For packages of seven beef snack sticks from a single processor, there were no significant differences in lethality between heating times, probably because of low trial numbers and relatively high intertrial variation. In initial trials with natural-casing wieners, increasing the heating time from 5.0 to 7.0 min resulted in significantly greater lethality (Table 2), whereas significant differences between wieners from different processors occurred with heating times of 6.0 and 7.0 min. In confirmatory trials (Table 3), there were no significant differences in lethality between beef snack sticks or natural-casing wieners from different processors. Throughout the study, there was noticeable intertrial variation. Potential causes of this variability include interlot variation in product composition, surface smoothness, and shape, variation in inoculum concentration, and variation in

TABLE 2. Initial trials to determine lethality of small-scale hot-water postpackaging pasteurization treatments against *Listeria monocytogenes* on commercial beef snack sticks and natural-casing wieners^a

Product	Treatment time (min)	Processor	Decrease in <i>L. monocytogenes</i> (log CFU/sample) ^b				Group mean
			Trial 1	Trial 2	Trial 3	Mean	
Beef snack sticks							
1/package	0.5	A	0.7	2.3 ^c	0.3	1.1	1.9 J
		B	2.4	1.7	1.3	1.8 A ^d	
		C	2.5	3.5 ^c	2.9	3.0	
	1.0	A	1.0	2.3 ^c	2.7 ^c	2.0	2.8
		B	3.7 ^c	2.5	2.9 ^c	3.0	
		C	3.6 ^c	3.2	3.6	3.5	
	1.5	A	4.4 ^c	2.3 ^c	2.7 ^c	3.1	3.4 J
		B	3.7 ^c	3.5 ^c	2.9 ^c	3.4 A	
		C	3.6 ^c	3.5 ^c	3.9 ^c	3.7	
4/package	3	B	1.4	1.5	0.2	1.0	1.1 KL
		C	1.4	1.4	0.9	1.2 B	
	4	A	3.9	3.9 ^c	4.0 ^c	3.9 C	2.9 KM
		B	2.7	3.9 ^c	1.4 ^c	2.7	
	5	A	4.0 ^c	3.9 ^c	4.0 ^c	4.0	4.0 LM
		C	2.1	1.9	2.5	2.2 BC	
7/package	3	B	2.5	2.7	1.5	2.2	1.8 N
		C	2.1	1.8	0.4	1.4	
	4	B	4.0 ^c	2.8 ^c	4.3 ^c	3.7	3.1 N
		C	2.2 ^c	2.8 ^c	2.8 ^c	2.6	
	5	A	2.3 ^c	2.1 ^c	4.1 ^c	2.8	2.7
		C	2.2 ^c	2.8 ^c	2.8 ^c	2.6	
	6	A	2.3 ^c	2.1 ^c	4.1 ^c	2.8	2.8
		C	2.2 ^c	2.8 ^c	2.8 ^c	2.6	
	Natural-casing wieners						
4/package	5	A	1.3	0.9		1.1	1.1 O
		B	1.0	1.0	1.1	1.0	
		D	1.3			1.3	
	6	A	2.0	1.2	1.5	1.6 DE	1.3
		B	1.1	1.1	1.4	1.2 DF	
		D	1.1	1.4		1.3 EF	
	7	A	4.3	1.1	1.9	2.4 GH	2.2 O
		B	1.4	3.4		2.4 GI	
		D	1.6	1.8		1.7 HI	
		C	1.6	1.8		1.7 HI	

^a In initial trials, no attempt was made to allow repair and enumeration of injured cells. A single package of product was submerged in 2.8 liters of boiling water for the time indicated and then chilled immediately in ice-water slush.

^b Each value is the result from an individual trial. Three treated samples were analyzed for each trial.

^c No surviving cells were detected. For such trial, a value of [log (zero time) - 1.9] was used for the decrease in cells.

^d Means within a column that have the same letter are significantly different ($P < 0.05$).

how closely the packaging film contacted the product surface.

Compliance guidelines from the USDA state that an effective postlethality treatment must reduce numbers of *L. monocytogenes* by at least 1.0 log unit (10). However, a higher standard of lethality must be met to reduce regulatory sampling frequency, which means *L. monocytogenes* numbers must be reduced by at least 2.0 log units. All of the PPP treatments tested in initial trials had some lethality against *L. monocytogenes* on vacuum-packaged beef snack sticks and natural-casing wieners (Table 2). With the compliance guidelines in mind, hot-water PPP treatments of at least 1.0 min for individually packaged beef snack sticks, at least 4.0 min for packages of four and seven sticks, and

at least 7.0 min for packages of four natural-casing wieners were viewed as having potential commercial utility. Some beef snack stick products do not support *L. monocytogenes* growth during storage (4). Application of validated hot-water PPP treatments to such products would allow the processor to operate under alternative 1 of the USDA regulations.

Treating individual beef snack sticks for 1.0 min often eliminated all inoculated *L. monocytogenes* cells. Results from these initial trials would support validation of the 1.0-min treatment for commercial use. Results of confirmatory trials using this treatment also support commercialization of this treatment (Table 3). Treating packages of four and seven beef snack sticks for at least 4.0 min also resulted in

TABLE 3. Confirmatory trials to determine lethality of small-scale hot-water postpackaging pasteurization treatments against *Listeria monocytogenes* on commercial beef snack sticks and natural-casing wieners^a

Product	Treatment time (min)	Processor	Decrease in <i>L. monocytogenes</i> (log CFU/sample) ^b				
			Trial 1	Trial 2	Trial 3	Mean	SD
Beef snack sticks							
1/package	1.0	A	3.6 ^c	3.5 ^c	3.4 ^c	3.5	0.1
		B	3.6 ^c	3.0 ^c	3.1 ^c	3.2	0.3
		C	2.7 ^c	4.4 ^c	4.5 ^c	3.9	1.0
4/package	4	A	3.6 ^c	3.8 ^c		3.7	
		C	3.4 ^c	3.1 ^c	3.0	3.2	0.2
7/package	4	A	2.9 ^c	3.2 ^c		3.1	
		B	3.3	1.9	3.1 ^c	2.8	0.7
		C	2.0	2.2 ^c	2.1	2.1	0.1
Natural-casing wieners							
4/package	7	A	2.0	1.6	1.8	1.8	0.2
		B	3.4	2.9	2.1	2.8	0.7
		D	1.8	2.0		1.9	

^a This procedure involved an attempt to allow repair and enumeration of injured cells. A single package of product was submerged in 2.8 liters of boiling water for the time indicated then chilled immediately in ice-water slush.

^b Each value is the result from an individual trial. One treated sample was analyzed for each trial. Standard deviation (SD) is given when $n = 3$.

^c No surviving cells were detected. For such a trial, a value of $[\log(\text{zero time}) - 1.9]$ was used for the decrease in cells.

reductions in *L. monocytogenes* concentration that met or exceeded the 2.0-log standard (Tables 2 and 3). Heating times needed to achieve a 2.0-log reduction increased with the number of pieces per package. The *L. monocytogenes* cells located at piece-to-piece contact surfaces could have been protected from the heating treatment. Results of trials to determine average surface temperature (Table 4) support this hypothesis.

Lethality of hot-water PPP against *L. monocytogenes* on natural-casing wieners was lower than that for beef snack sticks (Tables 2 and 3). This difference is likely re-

lated to the larger mass of the wieners compared with the beef snack sticks, which means that the wieners took longer to heat (Table 4). As with the beef snack sticks, piece-to-piece contact probably protected *L. monocytogenes* on the contact surfaces. A treatment time of 7.0 min appeared to result in sufficient lethality to meet the guideline for effectiveness but did not reach the lethality needed to reduce regulatory sampling frequency.

Comparison of Tables 2 and 3 suggests that greater reductions in numbers of *L. monocytogenes* occurred in confirmatory trials than in initial trials and that the overlay

TABLE 4. Surface temperature of beef snack sticks and natural-casing wieners during hot-water postpackaging pasteurization^a

Product	Processor	Surface temperature (°C) after:									
		0 min	0.5 min	1.0 min	1.5 min	2.0 min	3.0 min	4.0 min	5.0 min	6.0 min	7.0 min
Beef snack sticks											
1/package	A	5	43	47	56.5						
	B	5	42	51.5	56						
	C	5	43	49.5	51.5						
4/package	A	5		43.5		51	56.5	61	65		
	B	5		39.5		51.5	55.5	58	61		
	C	5		44		52	59.5	61.5	66		
7/package	A	5		39		51	59	61	62.5	67	
	B	5		40.5		46.5	52.5	58.5	62.5	66	
	C	5		42		49	55	62	66	67.5	
Natural-casing wieners											
4/package	A	5		42		45.5	53.5	58	58.5	62.5	62.5
	B	5		40		44	51.5	53	56.5	60	60.5
	D	5		39.5		45	47.5	54	58	60.5	63.5

^a Values are mean ($n = 2$) for average of three midpiece surface temperatures.

TABLE 5. Calculated reduction in *Listeria monocytogenes* during hot-water postpackaging pasteurization of beef snack sticks and natural-casing wieners^a

Product	Heating time (min)	Processor	F ₀ (min)	Reduction in <i>L. monocytogenes</i> (log CFU)	
Beef snack sticks					
1/package	0.5	A	0	0	
		B	0	0	
		C	0	0	
	1.0	A	0	0	
		B	0.01	0.01	
		C	0.01	0.01	
	1.5	A	0.05	0.04	
		B	0.05	0.04	
		C	0.02	0.02	
4/package	3.0	A	0.12	0.11	
		B	0.10	0.09	
		C	0.24	0.21	
	4.0	A	0.49	0.43	
		B	0.28	0.25	
		C	0.78	0.69	
	5.0	A	1.75	1.55	
		B	0.70	0.62	
		C	2.40	2.13	
	7/package	3.0	A	0.20	0.18
			B	0.04	0.03
			C	0.07	0.06
4.0		A	0.66	0.59	
		B	0.20	0.18	
		C	0.52	0.46	
5.0		A	1.42	1.26	
		B	0.81	0.72	
		C	2.20	1.95	
6.0		A	5.74	5.09	
		B	2.55	2.26	
		C	5.47	4.85	
Natural-casing wieners					
4/package	5.0	A	0.47	0.42	
		B	0.18	0.16	
		C	0.22	0.19	
	6.0	A	1.07	0.95	
		B	0.48	0.43	
		C	0.60	0.53	
	7.0	A	1.99	1.77	
		B	0.96	0.85	
		C	1.48	1.31	

^a Reductions were based on measured surface temperatures (Table 4) and the American Meat Institute Foundation process lethality spreadsheet with $D_{62.8} = 67.6$ and $z = 7.9^\circ\text{C}$.

plating procedure, which was intended to allow enumeration of injured cells, failed to achieve its objective. However, the overlay method always resulted in greater time-zero values than did direct plating in confirmatory trials. This finding indicates that some *L. monocytogenes* cells were initially injured upon exposure to the product surface. Numbers of *L. monocytogenes* cells recovered after the hot-water PPP treatments were similar for overlay and direct

TABLE 6. Cook-out loss during hot-water postpackaging pasteurization of beef snack sticks and natural casing wieners

Product	Treatment time (min)	Processor	% weight lost during cookout ^a	
Beef snack sticks				
4/package	3	A	0.9	
		B	0.2	
		C	0.8	
	4	A	0.9	
		B	0.6	
		C	0.6	
7/package	4	A	1.1	
		B	0.4	
		C	0.7	
	5	A	0.8	
		B	0.7	
		C	0.7	
Natural-casing wieners				
4/package	5	A	0.4	
		D	0.4	
		A	0.8	
	6	D	0.5	
		7	A	1.2
			B	1.0
	D		0.6	

^a Each value is for a single package of product.

plating methods, suggesting that few injured cells survived heating. The combination of greater time-zero cell numbers and comparable postheating cell numbers was responsible for the apparent difference in lethality between the initial and confirmatory trials.

Variability in results for wieners and beef snack sticks probably was due to slight differences in product size, product surface topography, heat transfer through adjoining pieces, and tightness of vacuum packaging. Increasing the PPP treatment time beyond those times studied here would undoubtedly decrease the variability of results and increase lethality but probably would not be economically feasible for processors.

The process lethality values calculated using surface temperature data are given in Table 5. Calculated process lethality values were generally much lower than those observed in initial (Table 2) and confirmatory (Table 3) trials, with the exception of packages of seven beef snack sticks heated for 6.0 min and packages of four natural-casing wieners heated 7.0 min. For these product-treatment combinations, calculated lethality was relatively closer to observed lethality. Possible reasons for the difference between observed and calculated lethality include compositional characteristics of beef snack sticks and natural-casing wieners that might enhance thermal destruction of *L. monocytogenes* relative to that occurring in the purge from smoked ham (the medium in which *L. monocytogenes* D - and z -values were determined), differences in thermotolerance between the strains used in the present study and those used by Muriana et al. (6), and lower than actual surface

TABLE 7. Consumer overall acceptance of beef snack sticks (seven per package) treated for 4.0 min with hot-water postpackaging pasteurization^a

Assigned descriptor	Numerical score	No. of responses for product from processor:					
		A		B		C	
		Trtmt	Ctrl	Trtmt	Ctrl	Trtmt	Ctrl
Like very much	7	64	65	52	49	76	66
Like moderately	6	71	69	84	66	61	89
Like slightly	5	33	35	32	33	41	23
Neither like nor dislike	4	6	7	8	11	7	7
Dislike slightly	3	5	4	7	18	4	6
Dislike moderately	2	2	2	2	7	2	1
Dislike very much	1	1	0	1	2	1	0
Mean		5.95	5.98	5.84	5.47	5.98	6.04
Trtmt significantly different?		NO		YES		NO	

^a Cooked-out fat and moisture were removed before presentation. Trtmt, treated product; Ctrl, untreated product.

temperature measurements resulting from product cooling during the 5 to 10 s that elapsed while the package was opened and the surface temperature was measured.

Several related studies on PPP of RTE meat products have been conducted. Collectively, these studies differ from the present one in three important ways: (i) the desired target for reducing numbers of *L. monocytogenes* cells was often greater than that required in the USDA compliance guideline, (ii) different products were tested, and (iii) the volume of hot water used for treatments and the degree of circulation and source of heat ensured that addition of cold packaged products did not cause any reduction in water temperature. Thus, direct comparisons between results of the present study and those of earlier work are difficult. Chen et al. (3) studied PPP of peeled (no casing) frankfurters (five per package) using 96°C water. The frankfurters also contained pediocin, which may have enhanced thermal lethality relative to that observed in our study. Reported reductions in *L. monocytogenes* were approximately 2.5 to 3.5 log CFU in 30 to 120 s. Murphy et al. (7) reported that a 5-min exposure of 4 kg of inoculated turkey breast to 96°C water in a cooker with a heat transfer coefficient of 800 W/m²K resulted in a 2.0-log reduction in

L. monocytogenes numbers. Similarly, Muriana et al. (5) found that a 2-min exposure of 1.8 to 5.0 kg of deli turkey product in 189 liters of 93.3°C water always resulted in a ≥2.0-log reduction in *L. monocytogenes* numbers.

Although process conditions in the present study were very specific, certain adjustments could be legitimately made to increase processing versatility without losing lethality. For example, the same product and packaging material could be processed using larger volumes of boiling water, salt in the water to elevate the boiling temperature, and/or increased heat source power. However, changing the product composition, number of product pieces per package, or type of packaging film would invalidate application of the present study results.

In addition to the challenges inherent in achieving consistently adequate lethality in an economical manner, processors adopting PPP would face the challenge of maintaining desirable sensory properties in the treated product. The cook-out loss in product treated by PPP ranged from 0.2 to 1.1% by weight for beef snack sticks and from 0.4 to 1.2% by weight for natural-casing wieners (Table 6). Cook-out loss could potentially be reduced by the addition of phosphates to the products. Processors seeking to use

TABLE 8. Consumer overall acceptance of natural-casing wieners (four per package) treated for 7.0 min with hot-water postpackaging pasteurization^a

Assigned descriptor	Numerical score	No. of responses for product from processor:					
		A		B		C	
		Trtmt	Ctrl	Trtmt	Ctrl	Trtmt	Ctrl
Like very much	7	53	60	66	45	56	45
Like moderately	6	79	81	68	79	71	76
Like slightly	5	35	24	29	26	33	35
Neither like nor dislike	4	5	9	12	14	7	12
Dislike slightly	3	6	6	6	10	9	7
Dislike moderately	2	3	3	2	6	2	3
Dislike very much	1	2	0	1	4	0	0
Mean		5.83	5.93	5.90	5.55	5.85	5.74
Trtmt significantly different?		NO		YES		NO	

^a Product was cooked in boiling water before presentation. Trtmt, treated product; Ctrl, untreated product.

TABLE 9. Consumer acceptance of the in-package appearance of beef snack sticks (seven per package) treated for 4.0 min and natural-casing wieners (four per package) treated for 7.0 min with hot-water postpackaging pasteurization^a

Assigned descriptor	Numerical score	No. of responses			
		Beef snack sticks (processor B)		Natural-casing wieners (processor B)	
		Trtmt	Ctrl	Trtmt	Ctrl
Extremely appealing	7	13	36	34	23
Moderately appealing	6	36	79	92	63
Slightly appealing	5	39	44	39	45
Neither appealing nor unappealing	4	31	16	15	24
Slightly unappealing	3	51	12	15	33
Moderately unappealing	2	19	7	1	6
Extremely unappealing	1	9	4	2	4
Mean		4.17	5.37	5.52	4.92
Trtmt significantly different?		YES		YES	

^a Trtmt, treated product; Ctrl, untreated product.

hot-water PPP treatments may wish to investigate such formulation changes. When PPP-treated beef snack sticks (with cooked-out fat and moisture removed) were served alongside untreated beef snack sticks to untrained consumer panelists, no negative effect of the PPP treatment was detected (Table 7). The hot-water PPP-treated beef snack sticks from processor B were actually evaluated as significantly better than the untreated control. Similar results were obtained for the natural-casing wieners (Table 8). However, when cooked-out fat and moisture from beef snack sticks were clearly visible to panelists in follow-up sensory evaluation trials, the PPP-treated product was rated significantly less desirable in appearance than untreated product (Table 9). In contrast, the appearance of PPP-treated natural-casing wieners (with no product heating after the package was opened) was rated significantly better than that of untreated wieners. Panelist comments suggested that product color and shape (resulting from compression as the packaging film shrank during PPP) were important factors resulting in higher appearance scores. None of the comments indicated a problem with the amount of cooked-out fat and moisture.

It is possible to meet USDA compliance guidelines for an effective postlethality treatment by treating beef snack sticks and natural-casing wieners with the relatively simple technique of small-scale hot-water PPP. Before PPP can be widely adopted by small and very small processors, however, further validation studies are necessary. Processors must determine whether hot-water PPP treatments would result in product characteristics that are acceptable to their customers.

ACKNOWLEDGMENTS

This project was funded in part by the U.S. Department of Agriculture Food Safety and Inspection Service (contract USDA-FSIS-C-26-

2003). M. D. DeVita was supported in part by the University of Wisconsin–Madison Medical Scholars Summer Research Program. The authors gratefully acknowledge Sungjoon Jang for assistance with sensory evaluation panels.

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