I. Project Title and ID Number:

Post-Process Pasteurization of Packaged, Ready-to-Eat products for Control of *Listeria monocytogenes*

FINAL REPORT

NPB Project #: 01-110

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II. Abstract

The efficacy of a saturated steam-based post-process pasteurization system to reduce/eliminate *L. monocytogenes* on frankfurters and restructured hams was evaluated. Frankfurters were packaged in a single layer format (4 per package), rinsed with lactic acid (2 or 4%) and pasteurized to end point surface temperatures of 160, 170 and 180°F. In addition, frankfurters were individually packaged and treated with lactic acid (2 or 4%), vacuum packaged and pasteurized to end-point surface temperatures of 160, 170 and 180°F. Similarly, restructured hams (half hams) were treated with lactic acid (2 or 4%), vacuum packaged and pasteurized for 2 or 4 min.

Pasteurization of inoculated single layer franks to target surface end point temperatures of 160, 170 and 180 °F resulted in *L. monocytogenes* reductions ($p \leq 0.05$) of 0.92, 1.44 and 2.89 log CFU/frank, respectively. Treatment of the franks with lactic acid at either 2 or 4 % did not result in greater ($p > 0.05$) *L. monocytogenes* reductions. Greater reductions in *L. monocytogenes* reductions were observed when the frankfurters were individually packaged, with 2.33, 4.63 and 6.52 log CFU/Frank reductions at target surface end point temperatures of 160, 170 and 180 °F.

Pasteurization of hams (half) for 2 and 4 min resulted in *L. monocytogenes* reductions ($p \leq 0.05$) of 2.03 and 4.14 log CFU/Sq. Cm., respectively. Lactic acid treatment at either 2 or 4 % did not result in greater ($p > 0.05$) *L. monocytogenes* reductions.

Surface pasteurization of frankfurters resulted in minimal, but statistically significant increases ($p \leq 0.05$) in surface hardness values and lower L* values compared to the non-

treated controls. However, the a* and b* values were not affected by heat treatments. Pasteurization of hams did not result in changes ($p>0.05$) in the color values (L*, a* and b*) of the ham surfaces.

Post process pasteurization of frankfurters (in-package) and hams using saturated steam based Stork-RMS Protecon system is effective in reducing the risk of *L. monocytogenes*.

III. Introduction

Listeria monocytogenes has been recognized as an animal and human pathogen for over seven decades, and emerged as an important and deadly foodborne pathogen in the late 1980's (Johnson et al., 1990). The recent resurgence of *L. monocytogenes* as a processed meat-borne pathogen has been attributed to "evolutionary" processing techniques and ingredient changes that may have contributed to its higher incidence and survival in RTE meat products (Borchert, 1999).

The documented efficiency of thermal processing protocols to eliminate/reduce the risk of *L. monocytogenes* pathogen in processed meats (Wilson, 1989; WHO, 1988) suggest that the key issue with safety of cooked RTE products is post-process re-contamination. This is of great consequence in RTE meat products, as these products are not always subjected to heating and further cooking at point of consumption.

Two possible avenues to ensure that RTE meats are free of *L. monocytogenes* are to (i) aseptically process and package RTE meat products under strict sanitation, and (ii) surface pasteurize the unsliced meat and meat products (in-package) to eliminate the pathogen. Post-process pasteurization of RTE meats holds promise as this would ensure that the products, even if contaminated after processing would be safe. However, the degree and method of thermal treatment should be designed carefully to afford a level of safety as required (after risk assessment), and at the same time conserve the sensory characteristics of the product in terms of palatability and appearance.

IV. Obejctives

The objective of the study were to evaluate the effect of lactic acid (2 and 4 %) in combination with post-process pasteurization (Stork-RMS Protecon system) on *Listeria monocytogenes* reductions on frankfurters (single layer and individually packaged formats) and restructured hams.

V. Procedures

Product preparation: Frankfurters (beef, pork and turkey; eight per pound) and hams (half) were obtained from a local retail store and stored at 4° C until treatment and pasteurization.

Inoculum preparation: A five strain cocktail of *Listeria monocytogenes* [108 M, 109, serotype 4c ATCC, serotype 3 ATCC, and H7738 (food outbreak strain)] was used. The cultures were maintained on Tryptic Soy Agar (TSA; Difco, Detroit, MI) slants at 4°C. Fresh cultures of the inoculum were prepared from the slants by transferring the cultures to 5 mL of Tryptic Soy Broth (TSB; Difco, Detroit, MI) and incubating at 35°C for 24 hours. After incubation, 1 mL of fresh culture was transferred into 100 mL TSB centrifuge bottles and further incubated at 35 °C for 18 hours. Cultures were then centrifuged at 10,000 rpm for 10 minutes at 4°C (Beckman J2-21 M/E centrifuge, JA-14 rotor, Palo Alto, CA). Cultures were then resuspended with 50 mL of 0.1% peptone water (PW; Difco, Detroit, MI) and centrifuged for a second time at conditions previously stated. The remaining pellet was resuspended with 10 mL PW. All strains

were combined aseptically in a sterile bottle to form a 5 strain cocktail of *L. monocytogenes*.

Product inoculation: Frankfurter and ham packages were opened and individual surfaces were blotted dry with a paper towel. The products were placed on an oven tray and "mist" inoculated in a "bio-containment chamber". A 1 h attachment period was provided. Inoculated products were vacuum-packaged or acid rinsed individually (1 per package) or in a single layer (4 per package) format for franks and each ham separately, at the Kansas State University Aseptic Processing Laboratory in a Stork-RMS Protecon Post-Process Pasteurization System.

Acid treatment: Inoculated franks [except controls (no acid, no heat)] were acid treated using a spray washer developed by Kansas State University. The treatments tested were 2% and 4% lactic acid at 20 psi. For hams, the product was placed in a zip-loc bag and acid treated for 1 min, allowed to drip and vacuum packaged. The franks were aseptically vacuum-packaged and pasteurized to target product sub-surface temperatures of 160, 170 and 180 °F. The hams were pasteurized for 2 or 4 min in the pasteurizer. Temperature was measured in between the two middle frankfurters for single layer frankfurters, the slowest heating surface, and for the individually packaged frankfurters, sub-surface (1 mm from the surface) temperature was used as target temperatures. At a Stork-RMS Protecon chamber temp of 203 °F, come uptime for surfaces of frankfurters to reach 160, 170, and 180 °F was 38, 58, and 96 sec, respectively. The franks were chilled in an ice water bath for 15 min before sampling. Pasteurization times of 4.25, 5.00, and 6.00 min were required for single layer franks to attain temperatures of 160, 170 and 180 °F at the junction of the two franks.

Sampling: The entire frank from the individual packaged product or one frank from the two middle franks in the single layer package were aseptically transferred to a filter stomacher bag. Surface samples of the hams were obtained by aseptically excising the surfaces, both the cut and the skin sections separately. Each sample was homogenized in a stomacher (Tekmar Co., Cincinnati, OH) with 50 mL of 0.1% sterile PW for 2 minutes. Samples were serially diluted using 9mL peptone blanks and plated on Modified Oxford Agar (MOX; Oxoid Ltd., Basingstoke, Hampshire, England) and Tryptose Phosphate Agar (TPA; Difco, Detroit, MI). Plates were incubated at 35° C for 48 hr. Colonies were counted and reported as $\log_{10}$ CFU/frank or $\log_{10}$ CFU/Sq. cm.

VI. Results

Single layer frankfurters:

Acid treatment alone of frankfurters resulted in ca. 0.7 log reductions in surface *L. monocytogenes* ($p \leq 0.05$) and no interaction between the acid treatment and target temperature were observed. Pasteurization of inoculated franks to target end-point temperatures of 160, 170 and 180 °F resulted in *L. monocytogenes* reductions of 0.92, 1.44 and 2.89 log CFU/frank (pooled data).

To attain the target surface temperatures (between the two middle franks, the slowest heating surface) of 160, 170 and 180 °F required exposure of the packages to 205 °F

chamber temperature for 4 min 14 sec, 5 min 4 sec, and 6 min and 2 sec, respectively in the Stork Post-process Pasteurizer.

Pasteurization of the frankfurters in the Stork Post-process pasteurization system affected the L* ($p \leq 0.05$) values of the franks, and not a* and b* values. However, acid treatment with 2% lactic acid did not affect the product color ($p > 0.05$). Pasteurization of single layer franks to end point temperatures of 160, 170 and 180 °F resulted in lowering the L* value to 56.7, 56.4 and 56.1, respectively from 58.2 for the non-treated control sample. Although differences in L* values were observed instrumentally, they may not be visually significant (<2 units difference observed). The a* and b* values of the franks were not affected by either the temperature or acid treatment, indicating that the pasteurization treatment alone or in combination could be used to reduce *L. monocytogenes* contamination on the frankfurters in single layer format.

Surface heat treatment of frankfurters in Stork Post-process pasteurization system resulted in increased hardness ($p \leq 0.05$) compared to the non-treated control by ca. 500 units. Acid treatment of the franks did not affect ($p > 0.05$) hardness of the frankfurters.

Although differences ($p \leq 0.05$) in color (L*) and hardness values were observed instrumentally in frankfurters subjected to post-process pasteurization, the differences were very low and may not adversely affect the sensory characteristics of the product. Sensory panel evaluations of treated franks is required for this determination.

Individually packaged frankfurters:

Pasteurization of franks to target surface end point temperatures of 160, 170 and 180 °F resulted in 1.98, 3.97 and 5.50 log CFU/frank reductions ($p \leq 0.05$) in *L. monocytogenes*, respectively. Acid treatment of the franks with lactic acid at 2 or 4% did not result in differences in *L. monocytogenes* ($p > 0.05$) populations on the surface of the franks. Larger reductions in *L. monocytogenes* on frankfurters were achieved using an individual frankfurter format compared to the single layer (touching franks) format and shorter pasteurization times were required to reduce *L. monocytogenes* populations.

To attain the target surface temperatures of 150, 160, 170 and 180 °F, exposure of the packages to 205 °F chamber temperature for 38, 58 and 96 sec, respectively in the Stork Post-process Pasteurizer was required.

Listeria monocytogenes is considered a fairly acid tolerant organism and can grow at pH 4.6 (Ryser and Marth, 1991). The antimicrobial action of organic acids is reported to be in order of increasing effectiveness, acetic>citric>lactic>malic acids (Porter 1950). Palumbo and Williams (1994) reported larger reductions with lactic acid compared to acetic acid for a 2 min exposure time, and a 1.33 log CFU/frank reduction upon exposure to 2% lactic acid for 5 min. The franks were exposed for a shorter time (<1 min) to the acid, using a spray system, which would be applicable to the meat industry, prior to thermal pasteurization in the present study. The authors suggested that incorporation of some type of organic acid treatment, especially lactic or acetic, could provide additional

safety for frankfurters and reduce the risk of *L. monocytogenes* growth in the event of contamination of the frankfurters during peeling and packaging operations.

Color differences (L*, a* and b* values) between the heat or acid treated samples and the non-treated control samples were not observed ($p>0.05$) for the frankfurters when pasteurized to surface temperatures of 160, 170 and 180 °F. This probably was due to the shorter treatment times compared to the frankfurters in the single layer format.

Restructured Hams:

Pasteurization of hams for 2 and 4 min in Stork-RMS Protecon Post-process pasteurization system resulted in 2.03 and 4.14 log CFU/Sq. Cm. reductions ($p\leq0.05$) in *L. monocytogenes*, respectively (Fig. 7). Acid treatment of the hams with lactic acid at 2 or 4% did not result in differences in *L. monocytogenes* ($p>0.05$) populations on the surface of the hams. Ham surface temperatures of ≥150 °F was attained within 15 s at chamber temperature of 205 °F.

Color differences (L*, a* and b* values; Fig. 8) between the heat or acid treated samples and the non-treated control samples were not observed ($p>0.05$) for the hams pasteurized for 2 and 4 min.

Although surface heat treatment of restructured hams in Stork Post-process pasteurization system showed significant differences in the model, the individual effects of exposure time and acid treatment were not significant ($p>0.05$). Increase in ham surface hardness was within 200 force units compared to the control, non-treated samples (Fig. 9). Similarly, acid treatment did not affect ($p>0.05$) hardness of the hams. Further, these minor differences or increases in hardness of the ham surfaces may not be of practical significance as these hams are often sliced thin for serving or subjected to further heat treatments before consumption.

Conclusions

The risk of *L. monocytogenes* on frankfurters and hams can be reduced by incorporation of processing steps such as post-process, in-package pasteurization to eliminate *Listeria monocytogenes* surface recontamination of RTE products. Stork-RMS Protecon steam based post-process pasteurization system alone or in combination with acid treatment is effective in reducing *L. monocytogenes* populations on surfaces of frankfurters and can be used as a critical control point in the manufacture of frankfurters and similar RTE meat products.

The process does not result in significant deterioration in quality due to secondary heat exposure of the RTE meat surfaces and could improve the shelf life of these products. The effectiveness of the system could be improved by incorporating bacteriostatic agents either as a topical application or as an ingredient in the RTE meat formulations to reduce the risk or recovery of the sub-lethally injured organisms during subsequent refrigerated storage during commercial distribution and by the consumer at home.

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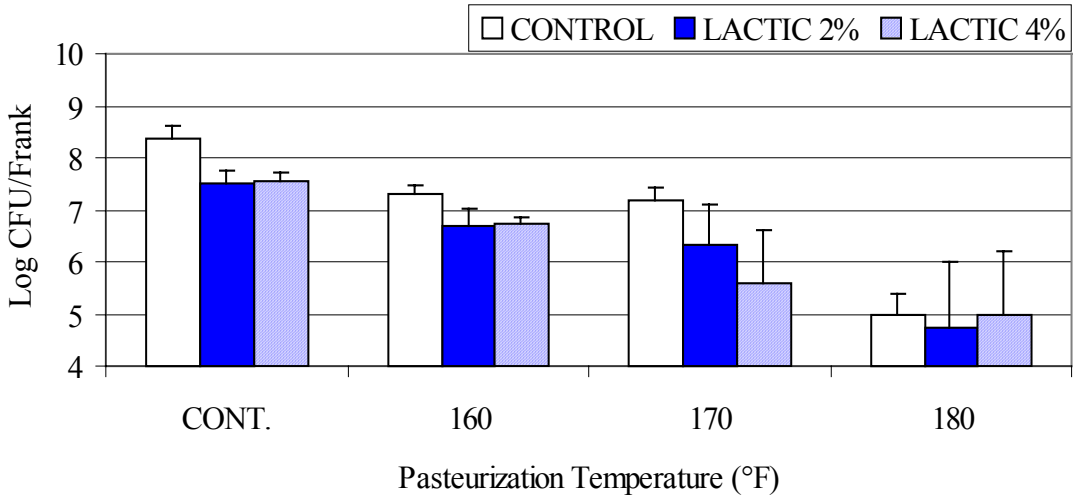

Fig. 1. *L. monocytogenes* populations on frankfurters (single layer format) surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

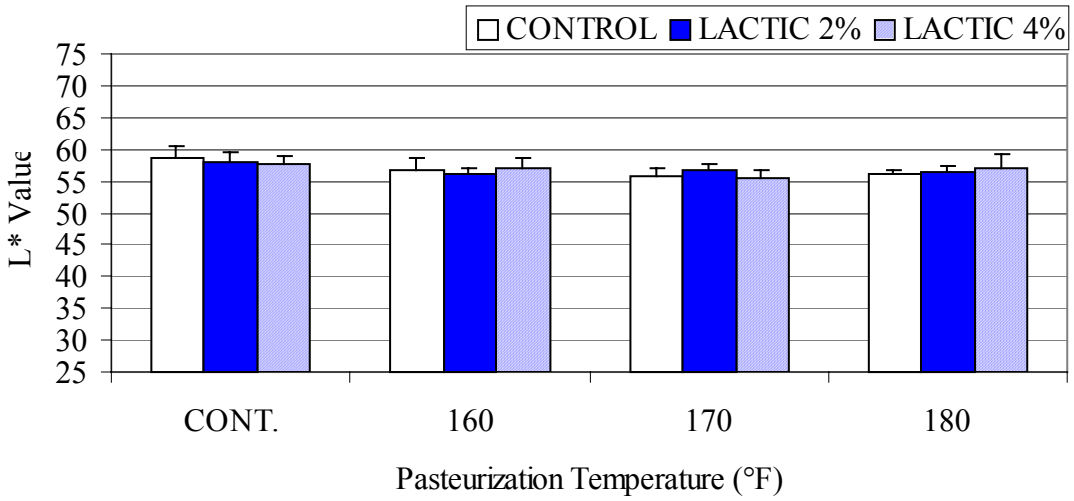

Fig. 2. Color (L*) values of frankfurters (single layer format) surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

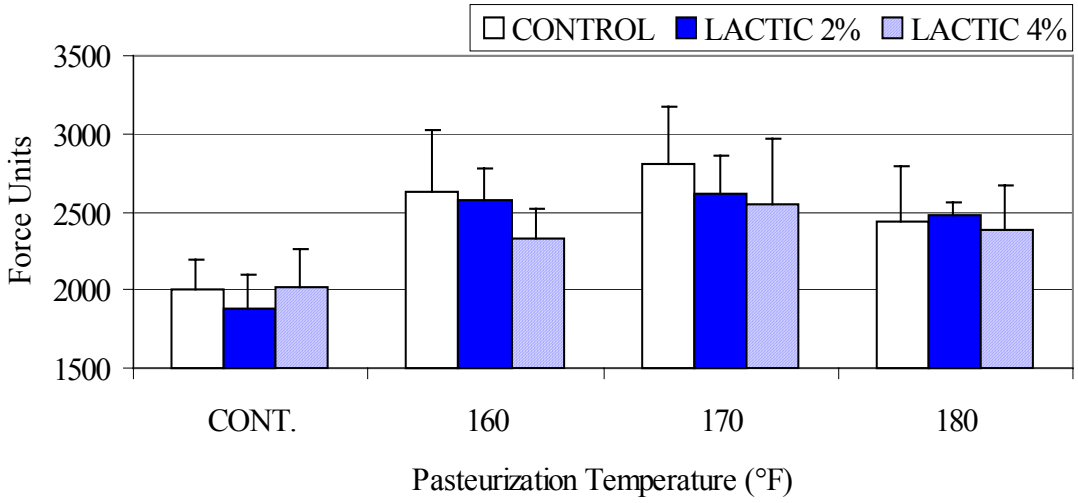

Fig. 3. Hardness of frankfurters (single layer format) surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

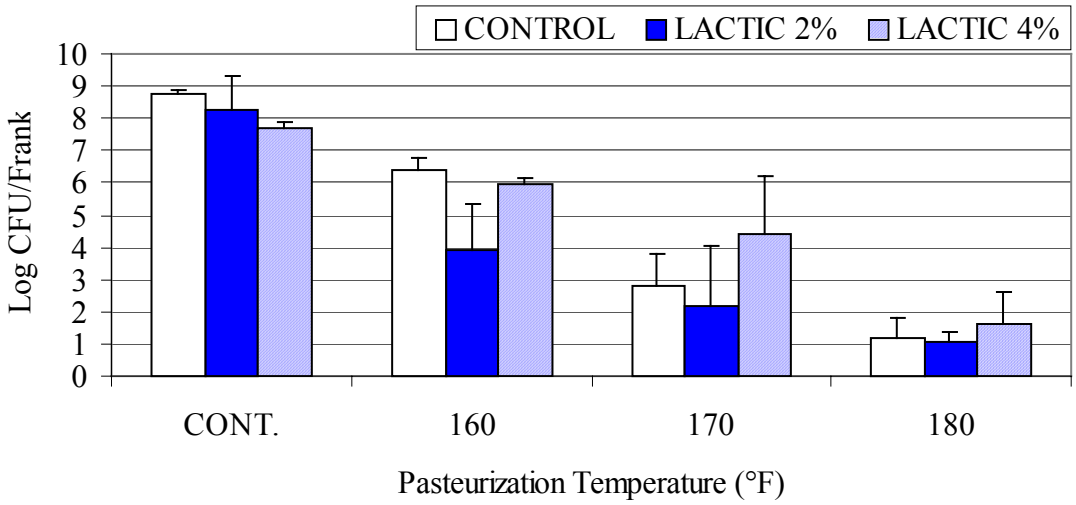

Fig. 4. *L. monocytogenes* populations on individually packaged frankfurters surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

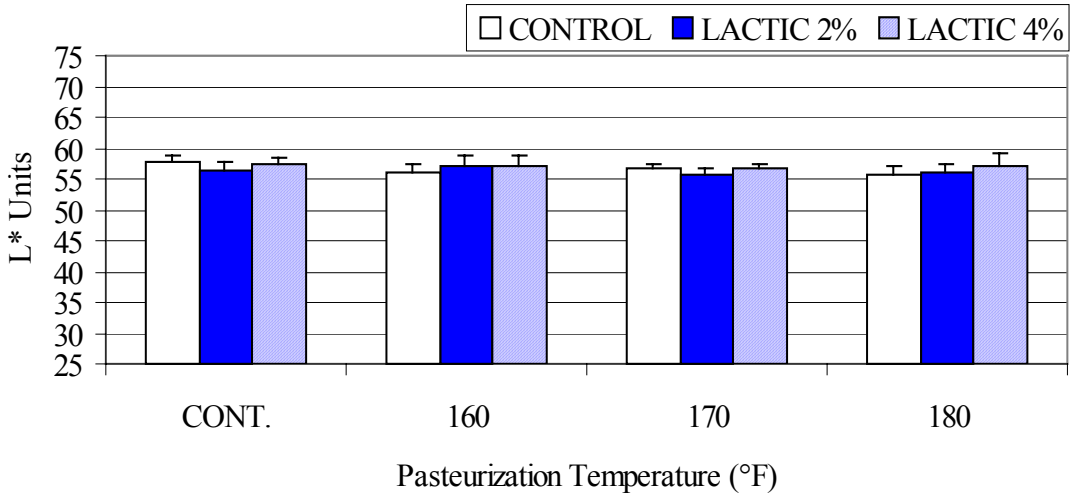

Fig. 5. Color (L*) values of individually packaged frankfurters surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

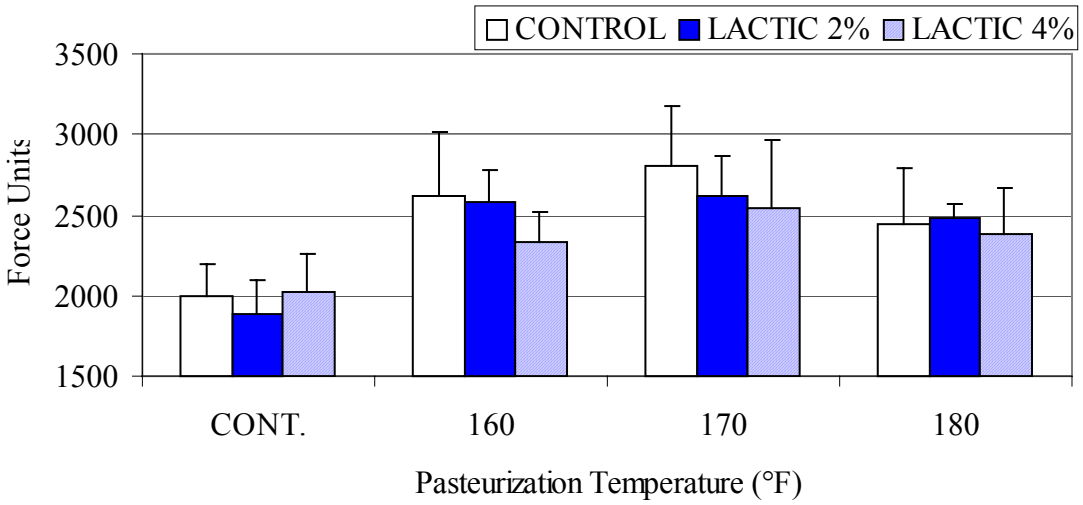

Fig. 6. Hardness of individually packaged frankfurters surface treated with lactic acid (2 and 4%) and subsequently pasteurized to 160, 170 and 180 °F in a Stork Post-process Pasteurization System.

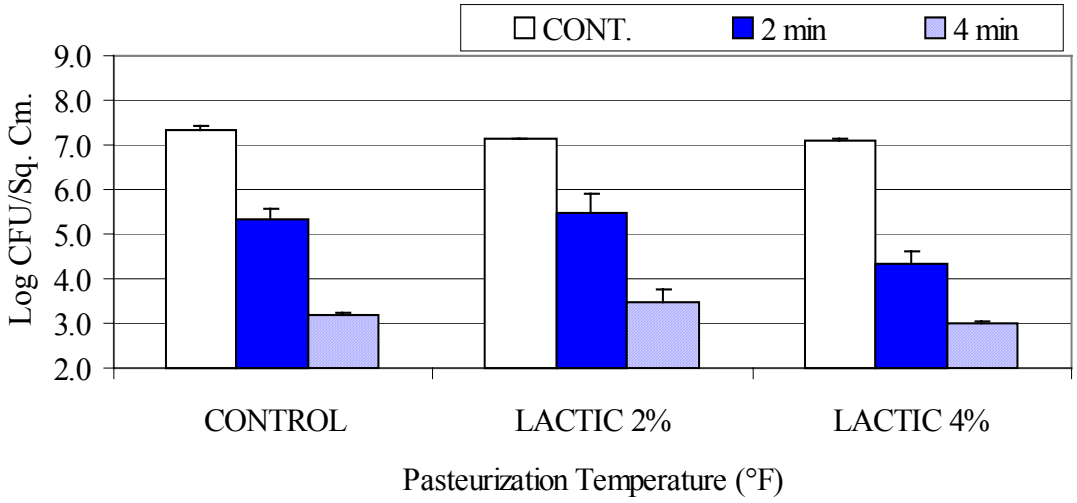

Fig. 7. *L. monocytogenes* populations on restructured hams (half) surface treated with lactic acid (2 and 4%) and subsequently pasteurized for 2 and 4 min in a Stork Post-process Pasteurization System.

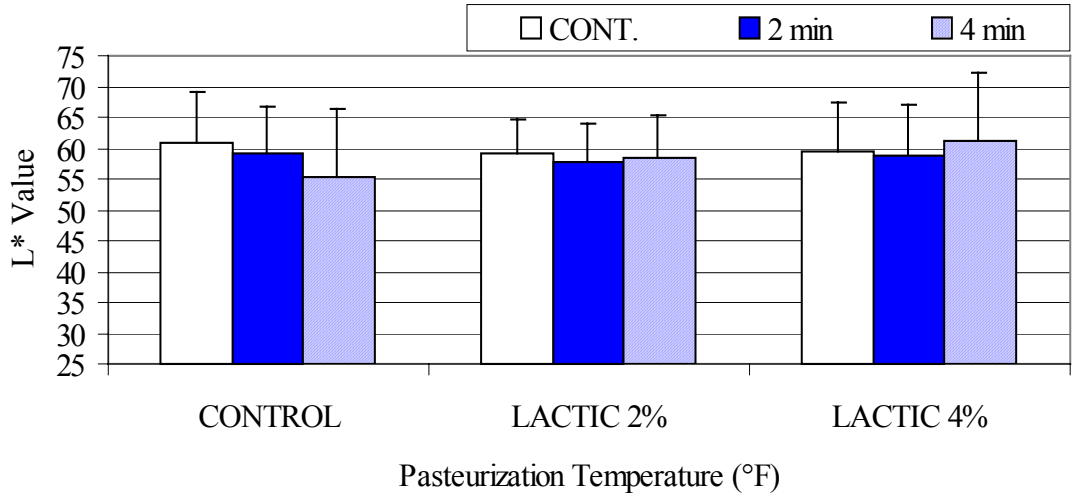

Fig. 8. Color (L*) values of restructured hams surface treated with lactic acid (2 and 4%) and subsequently pasteurized for 2 and 4 min in a Stork Post-process Pasteurization System.

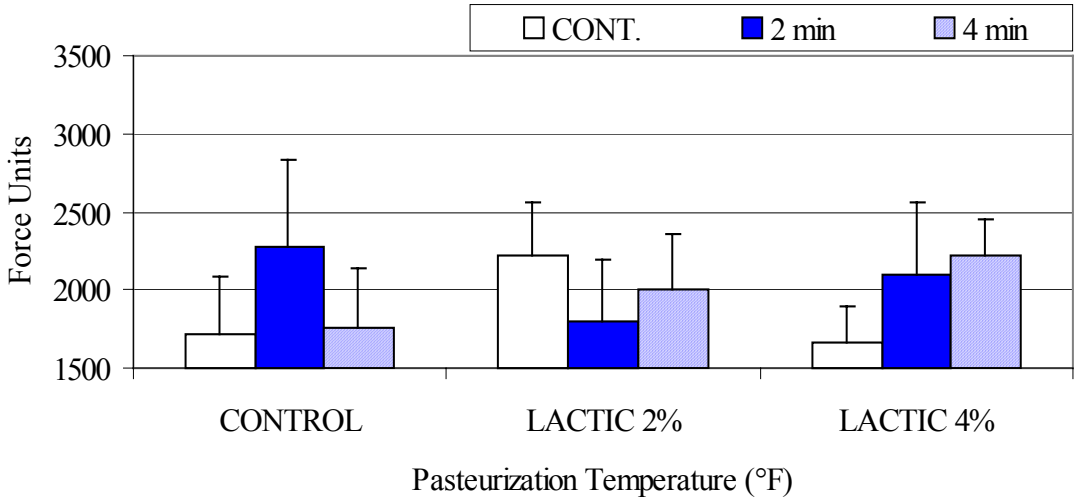

Fig. 9. Hardness of restructured hams surface treated with lactic acid (2 and 4%) and subsequently pasteurized for 2 and 4 min in a Stork Post-process Pasteurization System.