Research Note

Lactic Acid and Trisodium Phosphate Treatment of Lamb Breast To Reduce Bacterial Contamination

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ABSTRACT

Lactic acid and trisodium phosphate (TSP) were evaluated for the ability to reduce Escherichia coli and aerobic plate counts (APCs) on lamb breasts that were inoculated with a lamb fecal paste. A 90-s water rinse was applied followed by either a 9-s (55°C) 2% lactic acid spray, a 60-s (55°C) 12% TSP dip, or a combined treatment of both lactic acid and TSP treatments. Lactic acid reduced E. coli and APCs by 1.6 log_{10}/cm^2, and TSP caused a 1.8-log_{10}/cm^2 reduction in E. coli and a 0.7-log_{10}/cm^2 reduction in APCs. Combined reductions by the lactic acid spray followed by the TSP dip were 1.8 and 1.5 log_{10}/cm^2 for E. coli and APCs, respectively. Lactic acid and trisodium phosphate, used alone or in combination, were effective in reducing numbers of E. coli and could be useful as pathogen intervention steps in lamb slaughter processing.

Reducing or preventing carcass contamination during the slaughter process ensures a safer product will be produced. Factors such as the preslaughter condition of the animal and processing procedures affect carcass contamination (2). With respect to lamb, Biss and Hathaway (2) found that dirt and fecal matter present in the wool increased initial levels of microbiological contamination on the carcass.

Degree of contamination among sites on a lamb carcass varies (7, 14). Guthrie et al. (7) sampled lamb carcasses to assess coliform numbers. The highest coliform counts were found on the posterior pelvic rim, which is closest to the anus of the lamb where greater amounts of fecal material may be found. The ventral flank area contained intermediate levels, which is near the opening of the carcass for both pelt removal and evisceration, and the proximal hind limbs and the proximal forelimbs had the lowest counts. In reviewing these findings, coliform levels corresponded with the likelihood of contamination occurring at the pelt removal and evisceration. As outlined by Savell and Smith (16), care is stressed when removing the pelt from the anus and along the midline of the ventral area of the carcass and when eviscerating to prevent contaminating the carcass with ingesta or feces.

Lactic acid is an effective antimicrobial agent in reducing levels of contamination (1, 12, 18). Woolthuis and Smulders (20) used a 1.25% lactic acid spray on veal calf carcasses to determine the effects on microbial reduction. Aerobic plate counts (APCs) on the breast were reduced by 0.8 log_{10} and by 1.3 log_{10} on the perineum. These areas on a carcass traditionally have been major sites of contamination during evisceration. Greater reductions of bacteria were found as the concentration of lactic acid was increased (20). A 2.5-log_{10} reduction was observed when a 2% lactic acid concentration was applied to the carcass surface (20).

Trisodium phosphate (TSP) is an alkaline phosphate that has been used for the reduction of pathogens or other microorganisms on muscle foods (4, 8–11). Wang et al. (19) examined the effectiveness of a 10% (wt/vol) spray compared to that of a tapwater spray and 0.1% cetylpyridinium chloride spray. Counts of Salmonella Typhimurium were reduced by 0.7 to 1.6 log_{10} with tapwater spraying, were reduced by 1.6 to 2.3 log_{10} with the TSP treatment, and were reduced 1.5 to 2.5 log_{10} with the cetylpyridinium chloride application.

Investigating interventions such as lactic acid and TSP may prove beneficial to the lamb industry. Lactic acid does not require high-pressure applications, and the acid remains on the carcass allowing it to continue as an antimicrobial agent. The effectiveness of TSP in reducing pathogens appears to be a viable intervention. The industry may look into the combined uses of lactic acid and TSP as antimicrobial agents to reduce both pathogenic and aerobic bacteria.

This study provided the opportunity to test the ability of a 2% lactic acid (55°C) spray and a 12% TSP (55°C) dip in reducing Escherichia coli and APCs. It was possible to measure the performance of the treatments, both independently and jointly, that allowed an evaluation of the most effective intervention strategy available to reduce pathogens on lamb.

MATERIALS AND METHODS

Slaughter. Twenty-five lambs were slaughtered over a 3-day period at the Rosenthal Meat Science and Technology Center on...
the Texas A&M University campus in College Station following the procedures described by Savell and Smith (16). Only the breast region from each side of the carcass was utilized in this study, yielding 50 breast pieces. This region is described as being located from the 5th to 12th rib with a cut made no more than 4 in. from the lateral edge of the longissimus dorsi (17). The breast region was removed from the carcass during the slaughter procedure after evisceration and before final wash.

**Inoculation.** Fresh fecal matter was collected from lambs before slaughter and distributed into sterile stomacher bags in 10-g aliquots. Fecal samples were stored at refrigerated temperatures for no more than 24 h. Peptone diluents (9 ml of 0.1%; Difco, Detroit, Mich.) then were added to each bag and hand-kneaded to ensure a homogeneous, paste-like mixture. The fecal samples were spread over a 400-cm² area on each lamb breast using a sterile, stainless steel spatula.

**Application of interventions.** Each of the breast portions ($n = 50$) received a water rinse using a portable pump sprayer to remove the gross fecal matter from the carcass piece. The water rinse was applied in a light spray for 60 s followed by an increase in spray pressure for an additional 30 s. Following the water rinse, 25 of the breasts were treated with a 2% lactic acid spray at 55°C for 9 s (150 ml) using an insulated, portable pump sprayer. Following the lactic acid treatment, the breasts received a 12% TSP dip (20 liters) at 55°C for 1 min. The container used for holding the TSP at the required temperature was an Igloo cooler (Igloo Products Inc., Houston, Tex.). The remaining 25 breast regions, after the initial water rinse, received the TSP treatment only. Microbiological samples (procedures follow) were taken before and after these treatments to determine the efficacy of the individual and combined interventions.

**Determination of pH.** A portable, handheld pH meter, Jenco model 612 (Markson Science, Inc., Phoenix, Ariz.) with a flat bulb electrode (Markson Science, Inc.) was used to measure pH of the surface of the carcass piece before and after the application of the interventions. The pH was determined by placing the tip of the electrode flat against the surface of the carcass piece. Three readings were taken to provide a mean pH before and after the application of each intervention.

**Microbiological analysis.** Three representative surface samples measuring 10 cm² by 2 mm deep were excised from each lamb breast using a sterile, stainless-steel borer, sterile forceps, and a sterile scalpel. Between samples, the borer, forceps, and scalpel were sterilized by immersing in 95% ethanol followed by flaming. Surface samples were taken before inoculation to provide a background profile of natural microflora. Samples were obtained following inoculation to determine initial log counts before decontamination. All composites then were placed in a sterile stomacher bag with 100 ml of sterile 0.1% peptone and pummelled for 1 min using a Stomacher-400 (Tekmar Company, Cincinnati, Ohio). One milliliter of the pummelled composite was plated on both Petrifilm aerobic count (used to determine APCs) and $E. coli$ count plates. Appropriate 10-fold dilutions were plated in the same manner. Plates were incubated at room temperature for 48 h for APCs and 35°C for 24 h for $E. coli$ counts in the Texas A&M University Food Microbiology Lab. APC and $E. coli$ numbers were reported per cm².

**Statistical analysis.** One-way analysis of variance for mean reductions in log₁₀/cm² counts (APC and $E. coli$) with treatment (lactic acid, trisodium phosphate, and lactic acid plus trisodium phosphate) as the main effect was performed (15). When differences ($P < 0.05$) were found, mean separation was conducted using Tukey’s standardized test for significance between treatments (13).

### RESULTS AND DISCUSSION

The mean pH of the lamb breasts before treatment was 7.9. After treatment with lactic acid, the mean pH was 2.7, and after treatment with TSP, the mean pH was 12.3. The mean pH of the lactic acid plus TSP treatment was 12.5, which indicates that the basic effect of the TSP overpowered the acidic effect of the lactic acid spray.

Carcass samples were taken before inoculation in order to provide baseline counts of $E. coli$ and APCs. $E. coli$ counts for the three slaughter days averaged <0.1, 0.9, and 1.4 log₁₀/cm², respectively. APCs for the 3 days were 3.5, 3.5, and 3.8 log₁₀/cm², respectively. Samples also were obtained from the inoculated site before any applications of water rinses or interventions. Average inoculum levels for $E. coli$ were 5.7, 6.0, and 6.3 log₁₀/cm² for the 3 slaughter days, respectively, while mean APCs were 6.6, 6.6, and 7.0 log₁₀/cm², respectively. Mean counts by day did not differ ($P > 0.05$) (data not reported in tabular form).

Mean reductions for both APCs and $E. coli$ counts are reported in Table 1. For APCs, both the lactic acid and the lactic acid plus TSP treatments resulted in 1.6- and 1.5-log₁₀/cm² reductions from the control, respectively. The TSP treatment was less effective in reducing the APCs as evidenced by the lowest ($P < 0.05$) mean log₁₀/cm² reduction in counts compared to the control. For $E. coli$, all treatments were equally effective in reducing counts.

The effects of TSP on APCs in this study are similar to those findings reported by Dickson et al. (4), Kim and Slavik (8), Kochevar et al. (9), and Lillard (10). The application of lactic acid or TSP possesses benefits that outweigh those of spray washing. Ellerbroek et al. (5) sampled various areas on sheep carcasses in order to measure the effectiveness of spray washing. It was concluded that spray washing did not improve the microbial status of the sheep carcasses and that spray washing actually led to contamination of areas not likely to be otherwise contaminated. These findings are in agreement with Prasai et al. (13). The removal of such matter with a high-pressure sprayer will

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<tr>
<th>Table 1: Mean reductions by treatment for APC and $E. coli$ log₁₀/cm²</th>
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<tr>
<td><strong>APC</strong></td>
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<tr>
<td>Initial count, log₁₀/cm²</td>
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<tr>
<td><strong>Treatment</strong></td>
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<tr>
<td>Lactic acid, log₁₀/cm² reduction</td>
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<tr>
<td>Trisodium phosphate, log₁₀/cm² reduction</td>
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<td>Lactic acid/trisodium phosphate, log₁₀/cm² reduction</td>
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<td>MSD, log₁₀/cm² reduction</td>
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$^a$ Means within treatment section with different letters are significantly different ($P < 0.05$).

$^b$ MSD, Tukey’s minimum significant difference value for differences in treatment means.
either cause the contaminants to be spread elsewhere or cause the fecal material to be driven into the membrane (3).

Due to the nature of the fell membrane present on the lamb breast, high pressures could not be applied to remove contaminants. Contamination at levels used in this experiment would be rare in industry but were essential in determining the effectiveness of carcass interventions. Lactic acid and trisodium phosphate, used alone or in combination, were effective in reducing numbers of *E. coli* and could be useful as pathogen intervention steps in lamb slaughter processing. However, when gross contamination occurs, it is recommended that effective trimming be conducted to reduce visible contamination (6, 13) combined with a decontaminating lactic acid spray.

**ACKNOWLEDGMENT**

This research was supported, in part, by the Texas Food and Fibers Commission, Austin, Tex.

**REFERENCES**