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The Examination of Surface Contamination on Beef Carcasses During Slaughter and Aging in a Small-Scale Meat Packaging Operation Equipped with an Organic Acid Carcass Washer

[1]F. M. Steele and [2]D. Q. McMullin
[1]Department of Nutrition, Dietetics and Food Science,
Brigham Young University, Provo, Utah, USA 84602
[2]Novartis Pharmaceuticals, East Hanover, New Jersey, USA 07936

Abstract: Microbial contamination on beef carcasses was evaluated in a small-scale meat processing facility that was utilizing an organic acid wash system to minimize carcass contamination. Treatments were evaluated as a function of time in the process for aerobic plate count, lactic acid producing microbes, total coliforms and generic *Escherichia coli*. Treatments were, prior to the wash system, immediately after the wash and 24 h after the wash at-2 °C . Significant differences were observed between treatments for each class of microorganism. Sampling location (rump, fore shank and brisket) on the carcass was also evaluated and found to have a significant difference between locations ($\alpha = 0.05$). Results indicated significant reductions in beef carcass contaminants could be achieved through the carcass washing system being evaluated. Significant reduction of *E. coli* and total coliforms after the carcass wash was not observed due to the already low counts observed as a result of the wash itself.

Key words: beef carcass contamination, organic acid wash, sanitation, slaughter and aging

INTRODUCTION

Meat, poultry and fish are highly perishable foods. They are often the media through which food borne illnesses may spread. Although the majority of meat, poultry and fish borne illnesses are unarguably due to abusive treatment by the consumer, the level of contamination in the product due to the nature of its processing has continued to remain the focus of the public and governmental attention. It has long since been shown that the bacterial contamination of chicken increase during the cutting and packaging process (May, 1962). In a similar manner Cox *et al.* (1975) demonstrated that *Enterobacteriaciae* on poultry carcasses increase with processes such as continuous immersion chilling. It would be expected that similar relations could be identified with the processing of beef. In more recent years, however, meat packing plants have been instigating new sanitation techniques such as carcass washers with organic acid sprays or hot water rinses to aid in the reduction of the microbial flora immediately prior to chilling and aging (Gorman *et al.*, 1995; Graves *et al.*, 1997; Hardin *et al.*, 1995). Dickson and Anderson (1992) reviewed numerous investigations showing that such systems are successful in reducing total counts on carcasses. Similarly, Cutter and Siragusa (1994) demonstrated that organic acids in carcass washers are efficacious in reducing *Escherichia coli* O157:H7 contamination. These findings generate the question as to the state of the contamination of beef carcasses during processing which utilizes organic acid carcass washing technology. Most studies sited also utilize inoculated fecal material at the contaminating site on the carcass rather than evaluating the natural state of contamination typically present in the given slaughter facility as we chose to do (Gorman *et al.*, 1995; Graves *et al.*, 1997; Hardin *et al.*, 1995). The objective of this study, was to evaluate the efficacy of an organic acid washing system already established in a small-scale meat packing facility. The efficacy of the system would be tested on endogenous bacteria present on the beef carcasses as a result of the regular process operation of the facility, not from inoculated samples. Sampling sites and organisms of analysis were evaluated to determine the best method for determining best practices in overall sanitation in the carcass washing process. Samples were taken from the carcasses during regular production hours and followed through the process over time to the ageing of the carcass.

MATERIALS AND METHODS

Organic acid and spray wash treatment: A small-scale beef slaughtering operation (30-40 head day^{-1},

Corresponding Author: F. M. Steele, Novartis Pharmaceuticals, East Hanover, New Jersey, USA 07936

predominantly fed steers) was evaluated because of its use of an organic acid wash system. Final carcass washing was conducted in a two-chamber spray cabinet (CHAD, Lenexa, KS, USA). The first chamber consisted of a pressurized potable water (22 °C) rinse in a downward spray on the carcass, with a transit time of approximately 18 s. The second chamber consisted of a 2.5 % lactic acid solution rinse sprayed approximately an additional 18 s. Air curtains were operating at the end of each cabinet to minimize drip and aerosol transmission.

Sampling: Carcasses (n = 110) were randomly selected at varying times of the day and different days of the week for evaluation. Selected carcasses were sampled before (Treatment 1) and after (Treatment 2) the carcass wash treatment and again 24±2 h later after chilling in the aging cooler at-2 °C (Treatment 3). Each carcass evaluated was sampled in three areas most susceptible to post slaughter contamination. The areas sampled were the rump (Location 1), fore shank (Location 2) and brisket (Location 3) areas. Sampling consisted of two 5 cm^2 circles excised from the carcass using a sterile, stainless steel core sampler and then removing the core from the carcass aseptically with a sterile scalpel and tweezers. Samples were then placed in a sample bag, packed in ice and transported back to the laboratory. Samples were homogenized (Stomacher 400, Tekmar Co., Cincinnati, OH,USA) for 1 min in 100 mL 0.1% peptone buffer and then diluted serially to appropriate dilutions.

Microbiological analysis: Aerobic plate counts, total coliforms, E. coli and lactic acid bacteria were plated in duplicate using appropriate Petrifilm™ plates (3M Microbiology, St. Paul, MN, USA) according to AOAC international approved procedures (Curiale *et al*., 1991). Lactic acid bacteria were plated on Petrifilm™ aerobic count plates in an MRS diluent as a selective media and then incubated anaerobically at 32°C for 48 h according to 3M protocol for lactic acid bacteria testing in meats and meat products (Anonymous, 1998). CFU cm^{-2} were then calculated from the average of countable range plates.

Statistical analysis: Statistical analyses of the data were carried out by the General Linear Model (GLM) of the SPSS (1988). Where a statistically significant difference for a variable was detected, Tukey's HSD multiple range test was used to perform multiple comparisons of the levels of the variable. All statistical tests for bacterial counts were performed with log_{10} transformations. Where no detection of growth was obtained in the analyses, a value of 0.1 log_{10} was inserted which related to the sensitivity level of detection in the analyses

RESULTS AND DISCUSSION

In evaluating log_{10} bacterial counts across treatment groups, it is clear that the variances for *E. coli* and total coliforms were unequal (Fig. 1). The low counts obtained for these two analyses indicate that they are reduced

Table 1: Multiple comparison using Tukey's HSD determination for significance of treatment and sampling location means for each analytical variable with LOGCFU representing log_{10} values for aerobic plate counts, LOGMRS-log_{10} values for lactic acid producing bacteria, LOGCOLI-log_{10} values for total coliforms and LOGECOLI-log_{10} values for *Escherichia coli* counts

Dependent variable	(I) Treatment	(J) Location	Mean difference (I-J)	Std. Error	Sig.
LOGCFU	1.00	2.00	.4350*	0.1159	0.001
		3.00	.8906*	0.1022	<.0001
	2.00	1.00	-.4350*	0.1159	0.001
		3.00	.4556*	0.1222	0.001
	3.00	1.00	.4350*	0.1022	<.0001
		2.00	.8906*	0.1222	0.001
LOGMRS	1.00	2.00	1.0483*	0.1273	<.0001
		3.00	1.1565*	0.1123	<.0001
	2.00	1.00	-1.0483*	0.1273	<.0001
		3.00	0.1083	0.1342	0.699
	3.00	1.00	-1.1565*	0.1123	<.0001
		2.00	-0.1083	0.1342	0.699
LOGCOLI	1.00	2.00	.2441*	5.62E-02	<.0001
		3.00	.3252*	4.95E-02	<.0001
	2.00	1.00	-0.2441	5.62E-02	<.0001
		3.00	8.11E-02	5.92E-02	0.357
	3.00	1.00	-.3252*	4.95E-02	<.0001
		2.00	-8.11E-02	5.92E-02	0.357
LOGECOLI	1.00	2.00	0.111	4.84E-02	0.057
		3.00	.1641*	4.27E-02	<.0001
	2.00	1.00	-0.111	4.84E-02	0.057
		3.00	5.31E-02	5.11E-02	0.552
	3.00	1.00	-.1641*	4.27E-02	<.0001
		2.00	-5.31E-02	5.11E-02	0.552

Based on observed means, *. The mean difference is significant at the .05 level

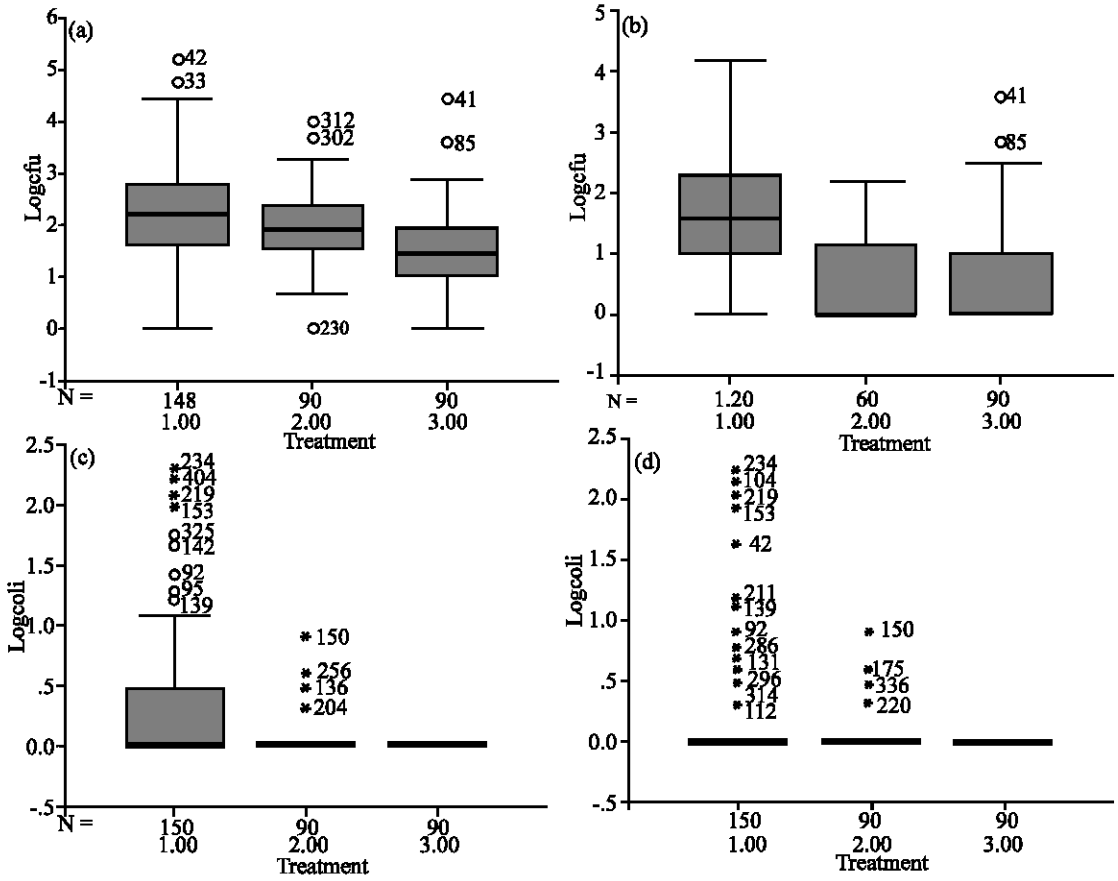

Fig. 1: Results for aerobic plate count (A), lactic acid producers (B), total coliforms(C) and Escherichia coli (D) across the three different treatment levels of 1.00 (before organic acid wash), 2.00 (immediately after organic acid wash) and 3.00 (24 h after acid wash stored at-2 °C). Results are illustrated as one box length representing the interquartile range (q3-q1). Outliers designated ° values--1.3-3 box lengths away from the median, * values--3+ box lengths away

by the organic acid wash and even more so by the cooling period. This may be an effect of the cooling, but more probable by the effect of the combination of cooling, time and the residual acid on the carcass. Tests show that there are significant differences across treatment, location and their interaction, but these outcomes are not sound because of their non-constant variance. By concentrating specifically on the differences within the treatment alone it is observed that there is statistical significance in the differences across the treatment levels. This can be seen in Table 1 as Tukey's HSD differentiates those differences between treatment levels. Consistent differences were observed between treatments 1 and 2, or in other words the effect of the lactic acid showed significant reduction in all dependent variable analyses. The effect of treatment 3, cooling for 24 h, showed significant reduction in aerobic plate count values, but did not significantly reduce counts of lactic acid producers, coliforms, or E. coli. This is probably relevant in the lactic acid producers, but probably not an issue with coliforms, or E. coli in that the counts of these two were already too low to be detected by our methods after the lactic acid wash, thereby making it difficult to achieve another significant reduction during the cooling period (treatment 3). This reduction in counts during the organic acid wash correlates well with the studies done by Anderson *et al.* (1987) using acetic acid and Hardin *et al.* (1995) using both acetic and lactic acids. These researchers did not, however evaluate their results from endogenous contamination, but from inoculated sample sites and, in the case of Hardin and others, they washed the samples in a model system and not in an existing carcass washing system. The differences observed across sampling location (Fig. 2), are less obvious even though they are significant at $\alpha = 0.05$. The differences seen here are of less consequence when evaluating the efficacy of a given

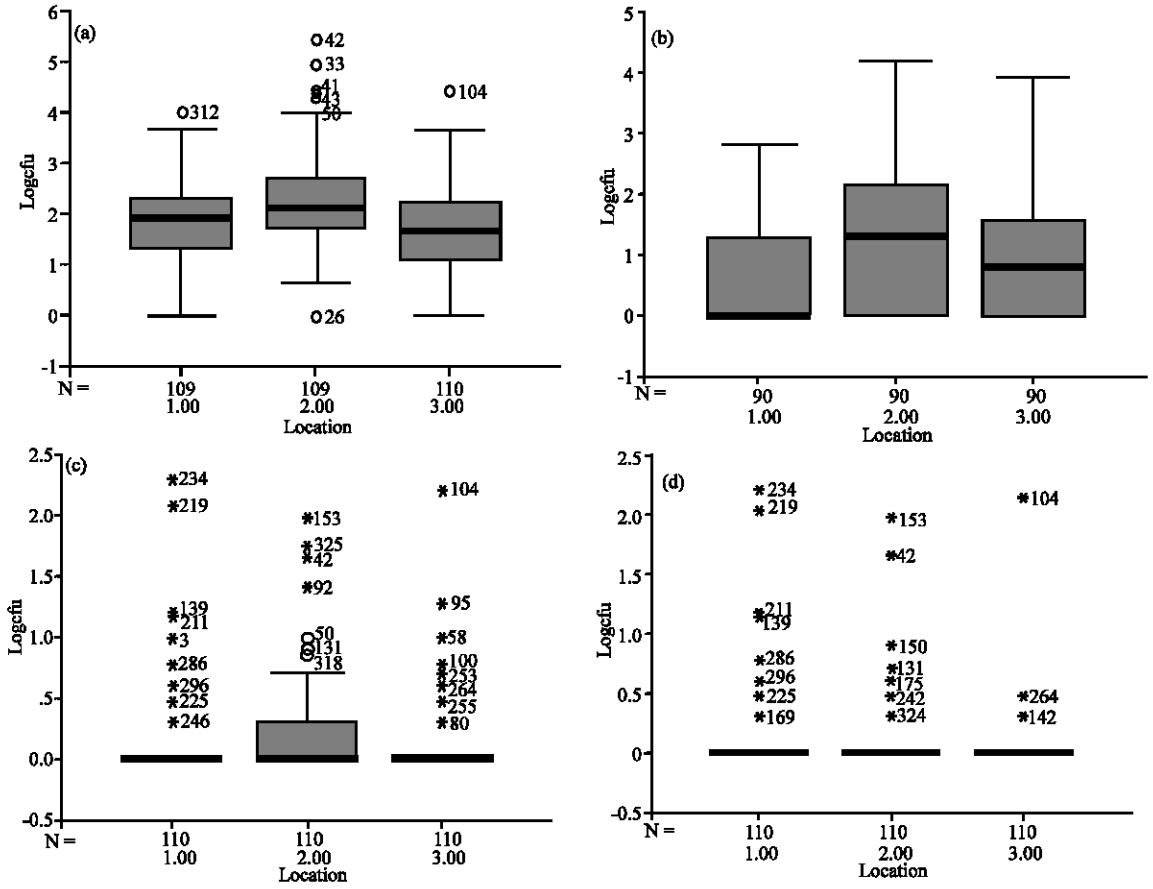

Fig. 2: Results for aerobic plate count (A), lactic acid producers (B), total coliforms(C) and Escherichia coli (D) across the three different locations sampled of 1.00 (rump), 2.00 (fore shank) and 3.00 (brisket). Results are illustrated as one box length representing the interquartile range (q3-q1). Outliers designated ° values--1.3-3 box lengths away from the median, * values--3+ box lengths away

treatment, but do show the importance of sampling location in the evaluation of carcass contamination. The three areas evaluated in this study are similar to those carcass sites evaluated in other studies (Dickson and Anderson, 1992; Jay, 2000) and typically represent areas that are most prone to contamination during the slaughter process.

Similar studies in this area of research have utilized a wide variety of methods to determine efficacy of a particular washing or sanitizing method. The ability of the methods used in detecting or determining microbial contamination or even ability to evaluate efficacy of a given treatment in meats and meat products is an issue important for meat processors. Enumeration of *E. coli* and even coliforms is usually so low that equal variance among the treatments is not possible as previously discussed and therefore make it difficult to get statistically significant results. These analyses do however, relate directly to the contaminating organisms of concern. Lactic acid bacterial counts give a good indication of spoilage microorganisms, but tend to be less useful due to the higher resistance of lactic acid producing microorganisms to the organic acid washing systems. The antimicrobial effect of organic acids is attributed to the undissociated ion of the acid and its ability to produce an environment less favorable for microbial growth (Hardin *et al*., 1995). It is also important to note that several researchers have shown that susceptibility of organisms to organic acid treatments , as shown in this study, varies with species (Davidson *et al*., 1973; Graves *et al*., 1997). Aerobic plate counts yielded the best statistical reliability because of the larger population numbers typically encountered, but give little direct relationship to primary organisms of concern. In conclusion, it is observed that a combination of several analyses (ie. coliforms, *E. coli* and APC) as well as specific presence/absence tests (i.e. Listeria,

Salmonella) be utilized to ensure a reliable microbiological evaluation of a given sanitizing system or process.

In general, these results demonstrated that it is efficacious in a small-scale facility to implement an organic acid wash system. The decreases observed in the plate counts of the organisms evaluated were significant to the point of justifying the system. It is also noteworthy, that the organisms present on the carcasses were organisms from the process and not inoculated from an optimized growth system. It is important to note that the implementation of an organic acid washing system at any scale slaughter facility, does not replace the need for good manufacturing practices throughout the facility to minimize the contamination of the carcass at any point in the process.

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