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Project Title: **Validation of Antimicrobial Interventions Including use of Peroxyacetic Acid (PAA) in a Bone Dust Cabinet and Spray Chill System in a Commercial Beef Harvest Operation**

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Objective: The objectives of this study were to evaluate the effectiveness of peroxyacetic acid (PAA) at two concentrations – low (180-220 ppm) and high (360-400 ppm) – for use in a bone dust cabinet against inoculated populations of non-pathogenic *Escherichia coli* biotype I, serving as surrogates for pathogenic *E. coli* and *Salmonella* spp., and to evaluate the effectiveness of the whole intervention system from harvest to the beginning of fabrication against the inoculated surrogates and beef carcass-associated natural microbial populations.

Experimental Design & Analysis:

The study was designed as a paired comparison replicated over two days with n = 40 or n = 60 samples per treatment (N = 560). For both treatments, carcasses were inoculated within 10 x 10 cm^2 marked zones. For treatment 1, uninoculated and inoculated A zones were sampled ("before treatment") and then carcasses were treated with low concentration PAA (180 to 220 ppm) in a bone dust cabinet and B zones were sampled ("after treatment"). Carcasses continued through remaining interventions (lactic acid and citric acid spray [LCA; 2.0% to 2.5%], PAA spray chill system [180 to 220 ppm], and post-chill lactic acid spray [LA; 1.5% to 5.0%]). After the post-chill LA spray, carcasses were sampled from both C zones ("after post-chill LA spray"). For treatment 2, uninoculated and inoculated zones were sampled ("before treatment") and carcasses were treated with a high concentration of PAA (360 to 400 ppm) in a bone dust cabinet, followed by a LCA spray (2.0% to 2.5%). Both B zones were sampled after treatment with PAA ("after treatment") and both C zones were sampled after the LCA spray ("after LCA spray").

Data were analyzed using the Mixed Procedure of SAS version 9.3. Orthogonal contrasts were used to determine significant treatment differences at similar points in the system as well as the differences between the whole system (after LA spray; treatment 1) versus the end of interventions on the harvest floor (after LCA spray; treatment 2).

Key Results:

Treatment 1, with the low concentration of PAA in the bone dust cabinet reduced ($P < 0.05$) initial inoculated counts by 2.2 log CFU/cm^2. For carcasses that received all the interventions in the whole system, EB counts were < 4.8 log CFU/cm^2 lower ($P < 0.05$) than those of carcasses that were sampled before the bone dust cabinet. Treatment 1, after the carcasses received the PAA treatment from the bone dust cabinet, uninoculated APC were reduced ($P < 0.05$) to < 0.1 log CFU cm^2. Carcasses that also received all the remaining interventions in the whole system, uninoculated APC were reduced by approximately <1.2 log CFU/cm^2. For Treatment 2, application of the high PAA concentration bone dust cabinet treatment reduced ($P < 0.05$) inoculated EB populations by 2.4 log CFU/cm^2. In addition to the PAA bone dust cabinet, the carcasses received the LCA spray treatment reduced ($P < 0.05$) EB populations by 3.0 log CFU/cm^2. For Treatment 2, a result of the PAA bone dust cabinet, uninoculated APC were reduced ($P < 0.05$) from 1.1 log CFU/cm^2 to < -0.3 log CFU/cm^2. Subsequently, carcasses received an LCA spray treatment and APC counts were reduced to < 0.3 log CFU/cm^2.

How can this information be applied in the industry?:

This could be applied to the beef industry as an alternative multiple-hurdle intervention system without the use of hot water.

Table 1. Least squares mean *Enterobacteriaceae* plate counts (log CFU/cm^2; [standard error]) for inoculated beef carcass zones before (untreated control) and after treatments (Treatment 1 and Treatment 2) with acid interventions (peroxyacetic acid [PAA], lactic acid [LA], and lactic acid plus citric acid blend [LCA]).

Treatment	Untreated Control	Bone Dust Cabinet Wash (low PAA vs. high PAA)*	Post Acid Treatments (Whole System Post-LA vs. Post-LCA Treatment)**
1- Whole System Interventions	5.8^a (0.2)	3.6^b (0.2)	< 1.0^c (0.2)
2- Bone Dust Wash (high PAA)	5.9^a (0.3)	3.5^b (0.3)	2.9^c (0.3)
Contrast *P*-Value	N/A	0.1851	<0.0001*

^{a,b,c} LSMeans bearing different superscript letters within the same row are different ($P < 0.05$)

LSMeans with a less than symbol (<) indicate at least one sample within the treatment had counts that were below the detection limit (-0.9 log CFU/cm^2)

P-values < 0.05 are considered different via orthogonal contrasts within each column

*PAA Bone dust (low = 180 to 220 ppm; high = 360 to 400 ppm)

**additional interventions include: LA = 1.5% to 5.0%, LCA = 2.0% to 2.5%

"N/A" Indicates that this *P*-value was not calculated

Table 2. Least squares mean aerobic plate counts (log CFU/cm^2; [standard error]) for uninoculated beef carcass zones before (untreated control) and after treatments (Treatment 1 and Treatment 2) with acid interventions (peroxyacetic acid [PAA], lactic acid [LA], and lactic acid plus citric acid blend [LCA]).

Treatment	Untreated Control	Bone Dust (low PAA vs. high PAA)*	Post Acid Treatments (Whole System Post-LA vs. Post-LCA)**
1- Whole System Interventions	1.0^a (0.2)	< 0.1^b (0.2)	< -0.2^c (0.1)
2- Bone Dust Wash (High PAA)	1.1^a (0.1)	< -0.3^b (0.1)	< 0.3^c (0.1)
Contrast P-Value	N/A	0.0009*	0.0061*

^{a,b,c} LSMeans bearing different superscript letters within the same row are different ($P < 0.05$)

LSMeans with a less than symbol (<) indicate at least one sample within the treatment had counts that were below the detection limit (-0.9 log CFU/cm^2)

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