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# Transportation and Lairage Environment Effects on Prevalence, Numbers, and Diversity of *Escherichia coli* O157:H7 on Hides and Carcasses of Beef Cattle at Processing†

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## ABSTRACT

Hide has been established as the main source of carcass contamination during cattle processing; therefore, it is crucial to minimize the amount of *Escherichia coli* O157:H7 on cattle hides before slaughter. Several potential sources of *E. coli* O157:H7 are encountered during transportation and in the lairage environment at beef-processing facilities that could increase the prevalence and numbers of *E. coli* O157:H7 on the hides of cattle. On three separate occasions, samples were obtained from cattle at the feedlot and again after cattle were stunned and exsanguinated at the processing plant (286 total animals). The prevalence of *E. coli* O157:H7 on hides increased from 50.3 to 94.4% between the time cattle were loaded onto tractor-trailers at the feedlot and the time hides were removed in the processing plant. Before transport, nine animals had *E. coli* O157:H7 in high numbers (>0.4 CFU/cm$^2$) on their hides. When sampled at the slaughter facility, the number of animals with high hide numbers had increased to 70. Overall, only 29% of the *E. coli* O157:H7 isolates collected postharvest (221 of 764) matched pulsed-field gel electrophoresis types collected before transport. The results of this study indicate that transport to and lairage at processing plants can lead to increases in the prevalence and degree of *E. coli* O157:H7 contamination on hides and the number of *E. coli* O157:H7 pulsed-field gel electrophoresis types associated with the animals. More study is needed to confirm the mechanism by which additional *E. coli* O157:H7 strains contaminate cattle hides during transport and lairage and to design interventions to prevent this contamination.

Recent work has revealed that the major source of beef carcass contamination is cattle hides *(4, 7, 8, 20).* In a study done by Barkocy-Gallagher et al. *(4),* the carcass contamination rates during processing could not be accounted for by the fecal prevalence of *Escherichia coli* O157:H7, but could be accounted for by hide prevalence. Bosilevac et al. *(7, 8)* and Nou et al. *(20)* extended these findings by demonstrating that antimicrobial interventions targeting cattle hides lead to drastic reductions in the rates of carcass contamination with *E. coli* O157:H7.

Because hide is the main source of carcass contamination, it is crucial to minimize the amount of *E. coli* O157:H7 on cattle hides before slaughter. Cattle can shed *E. coli* O157:H7 in their feces at concentrations as high as $10^6$ CFU/g *(24).* One such high shedder could contaminate the hides of many animals, especially in high-density situations like the transport truck or lairage environment. Few studies have addressed the effects of external contamination sources on *E. coli* O157:H7 hide prevalence during transport and lairage. Barham et al. *(3)* isolated *E. coli* O157:H7 from samples taken from truck trailers before cattle were loaded. It was not determined whether these isolates were found on the hides of cattle after transport or whether they were ever transferred to the carcasses. In a study of lairage pens, Collis et al. *(13)* used marker bacteria to demonstrate the potential for the spread of bacteria between lots of cattle via the lairage environment. In another study, lairage pen floors harbored *E. coli* O157:H7 from one day to the next *(26).*

Several preharvest intervention strategies based on vaccination, probiotics, and phage therapy have been designed in an attempt to reduce fecal shedding of *E. coli* O157:H7 in feedlot animals *(10, 16, 22, 28, 29).* The assumption has been made that a reduction in this fecal shedding by feedlot animals will lead to a reduced number of organisms on the hides of animals presented for processing and thereby a decrease in carcass contamination. However, there are several potential sources of *E. coli* O157:H7 contamination during the transport and lairage (e.g., animals shedding high numbers of *E. coli* O157:H7, trucks, lairage pens, alleys, chutes, and restrainers) that would seem to make further contamination likely *(2, 12).* In one study, genetically similar isolates were obtained from the lairage environment surfaces and from cattle hides during processing *(12),* but cattle hides were not sampled at the feedlot to determine whether the organisms in question were present before transport.

These issues raise the question, ‘‘Could effective feed-

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lot interventions be negated by contamination during transport and lairage?'' To address this question, a project was designed to measure *E. coli* O157:H7 prevalence and numbers before and after transport and lairage and to track isolates using pulsed-field gel electrophoresis (PFGE). The objectives of this work were (i) to determine whether transport and lairage environments affect the prevalence and number of *E. coli* O157:H7 on cattle hides and (ii) to track the sources of hide contamination during transport and lairage via molecular genotyping. Our hypothesis was that increased contamination of the hides will occur during transport and lairage and that the major source of this contamination is the animals themselves.

## MATERIALS AND METHODS

**Animals.** A total of 286 Charolais crossbred cattle (122 heifers and 164 steers) were followed from the feedlot to the processing plant in this study. Cattle were transported approximately 125 mi (201 km) to the processing plant on three separate occasions. Group 1 consisted of 81 steers that were loaded on two tractor-trailers the afternoon of 9 May 2005 and processed the next morning. Group 2 consisted of 66 heifers and 83 steers loaded onto four tractor-trailers on 31 May and processed the next morning. Group 3 was 56 heifers loaded onto one tractor-trailer and one gooseneck stock trailer on 20 June and processed the next morning. At the processing plant, the animals were treated as lots of commercial cattle. No special accommodations were made. The animals were maintained as single lots through processing and were not intermingled with other cattle. The test cattle could have come in contact with other cattle through the bars of the lairage pens.

**Loadout sampling.** Hide and fecal samples were collected at the feedlot immediately prior to loading cattle into trailers. Animals were restrained in a squeeze chute for sampling. Hide samples were collected with a sterile sponge (Nasco, Fort Atkinson, Wis.) premoistened with buffered peptone water (BPW; Becton Dickinson, Sparks, Md.) by swabbing an area of approximately 1,000 $cm^2$ behind the left shoulder, using five strokes (one motion back and forth constituted a stroke) for each side of the sponge. Fecal samples (10 g) were collected by rectal palpation. Hide and fecal samples were collected at approximately 1:00 p.m. on each loading day. Shortly after sampling, the cattle were loaded onto trailers and transported to the processing plant and unloaded. On the following day the cattle entered the processing facility at approximately 9:00 a.m.

**Truck samples.** Before the cattle were loaded, samples were obtained from the interior of each tractor-trailer. These samples are referred to hereinafter as truck samples. Four samples were collected from the interior of each truck, and two samples were collected from the gooseneck stock trailer. One sample was collected from each of the four main animal holding areas on the trucks. Truck samples were collected with a sterile sponge premoistened with 20 ml of BPW by swabbing an area of approximately 1,000 $cm^2$.

**In-plant sampling.** Hide and carcass samples were collected at the processing plant. Immediately after stunning and bleeding, hide samples were collected with a sterile sponge premoistened with 20 ml of BPW by swabbing an area of approximately 1,000 $cm^2$ in approximately the same location where the sample was collected at the feedlot. The lairage environment consists of the spaces encountered by the animals from unloading to stunning. Contamination of the hide could have occurred outside of the strict lairage environment following stunning as the animals were moved from the restrainer to the roll-out belt before their arrival at the hide sampling location. In this study, the lairage environment definition was broadened to include the restrainer and roll-out belt. Preevisceration carcass samples were obtained after the hide was opened but before application of any antimicrobial interventions. Carcass samples were collected with two sterile sponges premoistened with 20 ml of BPW. Each sponge was used to swab an area of approximately 4,000 $cm^2$, as previously described *(1)*.

**Bacterial enumeration.** *E. coli* O157:H7 from carcass, hide, fecal, and truck samples was enumerated using a spiral plater (Spiral Biotech, Norwood, Mass.) following a modified version of the protocol used by Robinson et al. *(25)*. For carcass, hide, and truck samples, the sponge sample was homogenized by hand massage, enrichment medium was added, and 250 μl of this solution was placed in a microfuge tube that was vortexed and then held for 3 min to allow the debris to settle. Following the settling period, 50 μl of sample was spiral plated onto plates of ntChromagar supplemented with novobiocin (20 mg/liter; Sigma, St. Louis, Mo.) and potassium tellurite (0.8 mg/liter; Sigma). Plates were incubated overnight at 37°C. Colonies were counted by visual inspection of the plates, and suspect colonies were confirmed as possessing the O157 antigen by latex agglutination. For enumerating from fecal samples, the enrichment medium (90 ml of tryptic soy broth [TSB; Difco, Becton Dickinson] with phosphate buffer [TSB+$PO_4$; 30 g of TSB, 2.31 g of $KH_2PO_4$, and 12.54 g of $K_2HPO_4$] per liter of solution) was added to the 10-g fecal sample, and the mixture was homogenized by hand massage. One milliliter of the sample mixture was removed to a microtube and vortexed. Bacteria were enumerated as described for hide samples. Limits of detection for the enumeration assay were 200 CFU/g and 0.4, 0.4, and 0.05 CFU/$cm^2$ for the fecal, hide, truck, and carcass samples, respectively.

**Sample processing for prevalence.** Samples were processed according to methods previously described with slight modifications *(5, 6)*. Carcass, hide, and truck sponge samples were enriched with 80 ml of TSB after the 250-μl aliquot for enumeration was removed. Fecal samples were enriched in the 90 ml of TSB+$PO_4$ used for enumeration dilution. The sample bags were incubated at 25°C for 2 h and then at 42°C for 6 h and then were held at 4°C overnight. Following incubation, the samples were processed by immunomagnetic separation, in which 1 ml from each enrichment was mixed with anti-O157 immunomagnetic beads (Invitrogen, Carlsbad, Calif.). Fifty microliters of the final bead-bacteria complexes was spread plated onto ntChromagar and onto sorbitol MacConkey agar (Difco, Becton Dickinson) supplemented with cefixime (0.05 mg/liter) and potassium tellurite (2.5 mg/liter; Invitrogen). All plates were incubated at 35 to 37°C for 18 to 20 h, and up to three suspect colonies were picked and tested by latex agglutination (DrySpot *E. coli* O157, Oxoid, Ogdensburg, N.Y.). A PCR assay was used to confirm that each isolate harbored genes for the O157 antigen, H7 flagella, and at least one of the Shiga toxins *(14)*. Isolates were maintained as frozen stocks for later use in strain typing by PFGE.

**PFGE.** *E. coli* O157 isolate fingerprints generated and analyzed in this study were based on PFGE separation of *Xba*I-digested genomic DNA, as described previously *(5)*; this is the method used by members of PulseNet (http://www.cdc.gov/ncidod/dbmd/pulsenet/pulsenet.htm). Pulsed-field gel certified agarose (SeaKem Gold Agarose) was obtained from Cambrex Bio Science Rockland, Inc. (Rockland, Maine). Tris-borate-EDTA running buffer and proteinase K were purchased from Sigma. *Xba*I

TABLE 1. E. coli *O157:H7 prevalence before and after cattle transport and lairage*

| Group | No. of samples collected | Feces pretransport | | Hides pretransport | | Hides postharvest | | Preevisceration carcasses | |
|---|---|---|---|---|---|---|---|---|---|
| | | Prevalence[a] | No. of high shedders[b] | Prevalence | No. of animals with high hide counts[c] | Prevalence | No. of animals with high hide counts | Prevalence | No. of carcasses with high counts[d] |
| 1 | 81 | 7 (8.6) | 4 | 23 (28.4) | 6 | 72 (88.9) | 16 | 2 (2.5) | 0 |
| 2 | 149 | 11 (7.4) | 2 | 109 (73.2) | 3 | 142 (95.3) | 44 | 20 (13.4) | 0 |
| 3 | 56 | 4 (7.1) | 2 | 12 (21.4) | 0 | 56 (100) | 10 | 6 (10.7) | 0 |
| Total | 286 | 22 (7.7) | 8 | 144 (50.3) | 9 | 270 (94.4) | 70 | 28 (9.8) | 0 |

[a] Number (%) of positive samples.
[b] Number of animals excreting *E. coli* O157 at or above 200 CFU/g feces.
[c] Number of animals with *E. coli* O157 on their hides at or above 0.4 CFU/cm$^2$ in the 1,000-cm$^2$ sample area.
[d] Number of carcasses harboring *E. coli* O157 at or above 0.05 CFU/cm$^2$ in the 8,000-cm$^2$ sample area.

was purchased from New England Biolabs (Beverly, Mass.). *Salmonella* serotype Braenderup strain H9812 was used as a control and for standardization of gels *(15).* Banding patterns were analyzed and comparisons were made using Bionumerics software (Applied Maths, Sint-Martens-Latem, Belgium) employing the Dice similarity coefficient in conjunction with the unweighted pair group method using arithmetic averages for clustering. Isolates were grouped into types that likely had the same origin based on fingerprint pattern similarities. Types were defined strictly as isolates that grouped together and had identical banding patterns.

## RESULTS

Even though the cattle were transported a relatively short distance and held in lairage for less than 1 day, the hide prevalence of *E. coli* O157:H7, the number of animals that harbored high numbers of *E. coli* O157:H7 on their hides, and the number of *E. coli* O157:H7 PFGE types associated with the animals increased on each occasion of transport and slaughter.

The fecal prevalence of *E. coli* O157:H7 prior to loading animals onto transport trucks was 7.7% (22 positive samples of 286 samples tested) for the three trials, with a range of 7.1 to 8.6% (Table 1). The hide prevalence before animals were loaded was 50.3% (144 of 286 samples), with a range of 21.4 to 73.2%. When the cattle hides were sampled during processing, the prevalence had increased to 94.4% (270 of 286 samples), with a range of 88.9 to 100%. The prevalence of *E. coli* O157:H7 on hides rose by an additional 60.5, 22.1, and 78.6% for groups 1, 2, and 3, respectively, between sampling the animals at the feedlot and sampling them again in the processing plant immediately before skinning. *E. coli* O157:H7 was detected on 9.8% of the carcasses (28 of 286 samples). Carcass contamination comes mainly from the hide *(7, 20).* This contamination probably is affected by several factors, including *E. coli* O157:H7 hide prevalence within a lot, *E. coli* O157 numbers on individual animals, the specific carcass dressing procedures employed, and the competency of the personnel applying the dressing procedures.

*E. coli* O157:H7 counts were obtained for fecal, hide, and carcass samples to determine the contamination of each animal. Animals were classified in the ''high'' category when *E. coli* O157:H7 counts were above the limits of detection for the enumeration assay. Therefore, animals were classified as high fecal shedders when they were excreting *E. coli* O157 at or above 200 CFU/g of feces. Eight animals were classified as high shedders during this study. For a high hide counts classification, animals had counts of 0.4 CFU/cm$^2$ of hide or greater for the 1,000 cm$^2$ of hide area sampled. Before transport, nine animals had high numbers of *E. coli* O157:H7 on their hides. When sampled again at the processing facility, the number of animals with high hide counts had increased to 70. The largest increase was recorded for group 2, where the number of animals with high hide counts of *E. coli* O157:H7 increased from 3 to 44. This group also had the highest carcass prevalence at 13.4%. *E. coli* O157:H7 was not detected in the enumeration assays for any of the carcasses; thus, all *E. coli* O157:H7–positive preevisceration carcasses had counts of $<0.05$ CFU/cm$^2$ of carcass area sampled.

Potential sources of *E. coli* O157:H7 associated with the increased prevalence and load of *E. coli* O157:H7 on animal hides were (i) the animals themselves, (ii) the trucks, and (iii) the lairage environment. The lairage environment was not sampled for this study, but the animal holding areas of the trucks were sampled before animals were loaded (Table 2). Seven trucks and a gooseneck stock trailer were used to transport the animals in this study. The visible cleanliness of the interiors varied greatly. The animal holding areas of some trucks were very clean, with only small pockets of fecal matter. Other trucks that had hauled cattle earlier in the day and were not washed out between loads were heavily soiled with feces. *E. coli* O157:H7 was detected in at least one sample from all seven of the trucks. One truck sample in each group had detectable numbers in the enumeration assay, indicating *E. coli* O157:H7 counts of $\geq$0.4 CFU/cm$^2$. A truck sample from group 2 had the highest *E. coli* O157:H7 count, at 2.8 CFU/cm$^2$.

In an attempt to track the source(s) of *E. coli* O157:H7 hide contamination during transport and lairage, 1,399 *E. coli* O157:H7 isolates from both pretransport and postharvest animals and from trucks were analyzed by *Xba*I PFGE. Isolate patterns were broken down into those associated with feedlot or trucks and those of unknown origin (assumed to be from the lairage environment). We have extensive data regarding molecular genotypes of *E. coli* O157:H7 associated with this feedlot and in particular the

TABLE 2. E. coli *O157:H7 prevalence and load for trucks and cattle transported*

| Truck no.[a] | No. of positive truck samples[b] | Highest *E. coli* count for truck samples (CFU/cm$^2$)[c] | No. of animals loaded | No. of high shedders[d] | Prevalence (%) | | |
|---|---|---|---|---|---|---|---|
| | | | | | Pretransport feces | Pretransport hide | Posttransport hide |
| Group 1 | | | | | | | |
| 1 | 3/4 | 0.4 | 40 | ND[e] | ND | ND | ND |
| 2 | 1/4 | <LOD | 41 | ND | ND | ND | ND |
| Total | 4/8 | — | 81 | 4 | 8 | 28 | 89 |
| Group 2 | | | | | | | |
| 1 | 3/4 | <LOD | 33 | 0 | 3 | 42 | 85 |
| 2 | 1/4 | <LOD | 33 | 0 | 0 | 64 | 100 |
| 3 | 2/4 | <LOD | 41 | 1 | 7 | 80 | 100 |
| 4 | 4/4 | 2.8 | 42 | 1 | 17 | 98 | 98 |
| Total | 10/16 | — | 149 | 2 | 7 | 73 | 95 |
| Group 3 | | | | | | | |
| 1 | 2/4 | 0.4 | 48 | 2 | 8 | 17 | 100 |
| 2 | 0/2 | <LOD | 8 | 0 | 0 | 50 | 100 |
| Total | 2/6 | — | 56 | 2 | 7 | 21 | 100 |

[a] All trucks were truck-trailer combinations except truck 2 for group 3, which was a gooseneck stock trailer.
[b] Number of positive samples/number of samples collected.
[c] LOD, limit of detection.
[d] Number of animals excreting *E. coli* O157 at or above 200 CFU/g feces.
[e] ND, not done. These animals were not tracked during transport.

cattle used for this study. Samples had been collected from these cattle for several months before initiation of this study, and PFGE data were compiled for the isolates (data not shown). Based on these data, the *E. coli* O157:H7 diversity at this feedlot was very low. From the three transport groups, 588 isolates were obtained from hide and fecal samples collected at the feedlot. Two major PFGE types combined to make up 97.4% of the total isolate genotypes (573 of 588 types) (Table 3). Eleven PFGE types represented the remaining 2.6% of the isolate genotypes (15 of 588 types). Because of the low diversity of PFGE types on the animals leaving the feedlot, identification of new *E. coli* O157:H7 contamination was much simpler.

*E. coli* O157:H7 was isolated from 16 of the 28 truck samples, with at least one positive sample coming from each truck. From the 16 positive samples, 44 *E. coli* O157:H7 isolates were obtained. These isolates consisted of 19 PGFE types. Three of these PFGE types were found in multiple trucks, and two types were found in separate sets of trucks. These trucks loaded cattle on the same days and were from the same commercial trucking company. The third set of trucks harboring isolates with identical PFGE types loaded cattle 2 weeks apart and were from different commercial trucking companies. Several trailers contained multiple PFGE types, with two trailers harboring isolates of four different types and one trailer having isolates of five different types.

At the processing plant, 764 *E. coli* O157:H7 isolates were recovered from the samples, with 684 and 80 isolates from the hide and carcass samples, respectively. Overall, only 29% of the isolates collected postharvest (221 of 764 isolates) had PFGE types that matched those of the isolates collected before transport (Table 4). The majority of postharvest isolates (69%, 524 of 764) could not be matched to any pretransport PFGE types and were assumed to have come from the lairage environment. The remaining 2% of postslaughter isolates (19 of 764 isolates) had PFGE patterns that matched those of isolates collected from the trucks.

The distribution of sources was not the same for the

TABLE 3. E. coli *O157:H7 PFGE types of hide and fecal isolates collected before transport of cattle*

| Group | Total no. of isolates | No. (%) of isolates matching feedlot PFGE type: | | |
|---|---|---|---|---|
| | | 1 | 2 | Other ($n$ = 11) |
| 1 | 83 | 76 (91.6) | 6 (7.2) | 1 (1.2) |
| 2 | 466 | 352 (75.5) | 100 (21.5) | 14 (3.0) |
| 3 | 39 | 0 | 39 (100) | 0 |
| Total | 588 | 428 (72.8) | 145 (24.7) | 15 (2.6) |

TABLE 4. E. coli *O157:H7 PFGE types of hide and carcass isolates collected after harvest of cattle*

| Group | Total no. of isolates | No. (%) of isolates matching PFGE types from: | | |
|---|---|---|---|---|
| | | Feedlot | Trucks | Unknown origin |
| 1 | 201 | 179 (89.1) | 1 (0.5) | 21 (10.4) |
| 2 | 393 | 24 (6.1) | 0 | 369 (93.9) |
| 3 | 170 | 18 (10.6) | 18 (10.6) | 134 (78.8) |
| Total | 764 | 221 (28.9) | 19 (2.5) | 524 (68.6) |

three groups. In group 1, the majority of postslaughter PFGE types (89%, 179 of 201) matched PFGE types previously found at the feedlot, whereas only 0.5 and 10% came from the trucks and unknown sources, respectively. The other two groups were vastly different from group 1, with 94 and 79% of the isolates types associated with unknown sources for groups 2 and 3, respectively. In group 3, 18 (11%) of the 170 isolates were of PFGE types identical to those of isolates from the trucks.

## DISCUSSION

Multiple hurdle postharvest interventions have been very effective in eliminating carcass contamination *(1, 11, 21).* However, to maintain process control, the amount of contamination coming into the plant, as determined either by prevalence or counts, must be kept below a defined limit. When contamination exceeds this limit, the interventions will be overwhelmed and carcass contamination will not be eliminated. The incoming *E. coli* O157:H7 contamination of most concern is that found on the hides of cattle entering the processing facility *(4, 5, 7, 20).* Recently, postharvest interventions have been focused on the hide in an attempt to keep contamination levels from overwhelming downstream antimicrobial interventions applied to the carcass *(7–9).* The efficacy of a hide-on carcass wash cabinet in reducing hide and carcass contamination during processing was evaluated by measuring its effects on hides and corresponding preevisceration carcasses *(8).* Hide samples were collected before entry into and after exit from the cabinet. The prevalence of *E. coli* O157 was reduced on hides (from 44 to 17%) and on preevisceration carcasses (from 17 to 2%) when the cabinet was in use. These results support decontamination of hides as an effective means of reducing pathogen contamination of cattle carcasses during processing.

Another focus has been on reduction of fecal prevalence of *E. coli* O157:H7 by using preharvest interventions, assuming they would result in reduced hide contamination and subsequently reduced carcass contamination *(10, 16, 22, 28, 29).* A concern associated with this approach is that it is not clear to what extent *E. coli* O157:H7 prevalence and contamination levels increase after the animals leave the feedlot. Because no preharvest intervention could be 100% effective *(22, 29),* a few animals may be excreting high numbers of *E. coli* O157:H7 during transport and lairage. In the high-density environments of transport and lairage, a few animals shedding high numbers of *E. coli* O157:H7 could contaminate the hides of several animals in that lot *(17, 18).* In the current study, a small number of the cattle shedding *E. coli* O157:H7 (8.6% in group 1) can lead to dramatic increases in hide prevalence and counts (from 28 to 89% and from 6 to 16 animals with high hide numbers) in a very short period (Table 1). McGee et al. *(19)* found that within 24 h of introducing animals known to be shedding *E. coli* O157:H7 the hides of cohort animals became contaminated, and at 48 h 66% of the penmates had *E. coli* O157:H7 on their hides. In another study performed using molecular tracking methods, direct hide-to-hide contamination was a viable route for the transmission of *E. coli* O157:H7 during transport and lairage *(27).*

The isolation of *E. coli* O157:H7 strains with new PFGE types from cattle hide samples following the transport and lairage process is not necessarily a cause for concern. The discovery of new PFGE types could be indicative of low-level contamination events where the animals were picking up new *E. coli* O157:H7 types but at numbers too low to be problematic in downstream processing. One indication that this scenario did not apply in this study was the high proportion of carcass isolates that were of PFGE types not matching any collected at the feedlot. Over 80% (67 of 80) of the carcass isolates did not match pretransport PFGE types; 1 matched a truck isolate type and the other 66 were of unknown origin. These findings indicate that contamination events are occurring with substantial magnitude.

Currently, there are no standard antimicrobial interventions applied to either transport vehicles or lairage environments. Therefore, cattle that have undergone successful preharvest interventions may easily become contaminated from transport and lairage surfaces. Upon arrival at the processing plant, cattle are unloaded and directed through common alleys to lairage pens. The lairage pens may have housed other animals earlier in the day and may or may not have been cleaned between lots. Upon exit from the lairage pens, the cattle are directed through more common alleys before entering the snake, a chute designed to place the cattle in single-file, and coming to the area for stunning and shackling. Every animal arriving at the plant will follow the same basic path; hence, transfer of bacteria between lots is possible. In several studies, the presence of *E. coli* O157:H7 on processing plant lairage environment surfaces has been documented. Rivera-Betancourt et al. *(23)* recovered *E. coli* O157:H7 from 13% of the samples collected from fence panels in the lairage pens of a commercial processing plant. They also reported *Salmonella* and *Listeria* prevalences of up to 52 and 40%, respectively, in samples obtained from lairage pen fence panels. Similarly, Small et al. *(26)* recovered *E. coli* O157:H7, *Salmonella,* and *Campylobacter* in cattle lairages; *E. coli* O157:H7 was present in 27.2% of the lairage samples. *E. coli* O157:H7 can persist in lairage environments for several days and through cleaning periods *(27).* Few previous studies have addressed the issue of transport trucks as a source of *E. coli* O157:H7. In a study in which samples were collected from trucks before cattle were loaded, 7% of the trailers harbored *E. coli* O157:H7 and 74% harbored *Salmonella (3).* Childs et al. *(12)* also sampled trailer interiors before cattle were shipped, and *E. coli* O157:H7 was isolated from 5% of these samples. These authors listed transport trailers as a potential source for *E. coli* O157:H7 hide contamination based on their identification of a hide isolate and an isolate from a trailer that were probably related, as determined by PFGE analysis. In the current study, *E. coli* O157:H7 isolates with identical PFGE patterns were isolated from trucks and from postharvest hide samples. Those PFGE patterns did not match any patterns from the isolates collected at the feedlot, thus implicating the trucks as a direct source of hide contamination.

When this project was designed, the hypothesis was

that increased contamination of the hides would occur during transport and lairage and that the major source of this contamination would be the animals themselves. The results from group 1 support this hypothesis, but results from groups 2 and 3 are only partially supportive. Although the prevalence and counts of *E. coli* O157:H7 both increased, the main source of the contamination was not the animals themselves. A large proportion of contamination came from an unknown source, believed to be the lairage environment. One possible reason for this finding is that the animals were housed overnight in the lairage pens. Typically, cattle arrive at the processing facility and are housed in the lairage pens for only 2 to 5 h before they enter the facility for processing. However, cattle that arrive late in the day may be held overnight and processed the following morning, as happened with the three groups in our experiment. The lairage environment is usually washed during the night when the plant is not in operation. Any cattle in the lairage pens during the wash procedure most likely come in contact with the spray and runoff when adjacent pens are washed down. This contact might account for the dominant presence of nonfeedlot bacteria on the animals' hides in such a short period. More study is needed to confirm the mechanism by which new *E. coli* O157:H7 genotypes contaminate cattle hides during transport and lairage and to develop antimicrobial interventions to prevent this contamination.

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