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Research Note

Prevalence Rates of *Escherichia coli* O157:H7 and *Salmonella* at Different Sampling Sites on Cattle Hides at a Feedlot and Processing Plant†

NORASAK KALCHAYANAND,* DAYNA M. BRICHTA-HARHAY, TERRANCE M. ARTHUR, JOSEPH M. BOSILEVAC, MICHAEL N. GUERINI, TOMMY L. WHEELER, STEVEN D. SHACKELFORD, AND MOHAMMAD KOOHMARAIE‡

U.S. Department of Agriculture, Agricultural Research Service, Roman L. Hruska U.S. Meat Animal Research Center, Clay Center, Nebraska 68933-0166, USA

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ABSTRACT

The prevalence rates of *Escherichia coli* O157:H7 and *Salmonella* at different sampling sites on cattle hides were determined at a feedlot and a processing plant. Sponge samples were collected from six hide surface sites at the feedlot (left and right shoulders, left and right ribs, back, and belly) and four sites at the processing plant (left and right shoulders, back, and belly). The prevalence of *E. coli* O157:H7 was approximately 80% for left and right shoulder and rib samples, 68% for back samples, and 92% for belly samples collected at the feedlot. At the processing plant, the prevalences of *E. coli* O157:H7 at all four sites were between 76 and 79%. *Salmonella* prevalence in feedlot samples was too low to allow for accurate analysis. The prevalence of *Salmonella* at processing was 49% for left shoulder samples, 48% for right shoulder samples, 40% for back samples, and 68% for belly samples. The results of this study indicate that the site most likely to be naturally contaminated with *E. coli* O157:H7 and *Salmonella* simultaneously was the belly.

Escherichia coli O157:H7 and *Salmonella* are causative agents for foodborne illnesses. These two pathogens alone caused approximately 1.4 million foodborne illnesses and 600 deaths in the United States in 2000, with an estimated $3 billion in associated medical costs, productivity losses, and costs of premature deaths *(1)*. These pathogens pose both a significant health risk and a considerable economic threat to the beef industry.

Pre- and postharvest interventions to control or eliminate these pathogens should be an important part of animal husbandry practices *(10)*. However, the prevalence of *E. coli* O157:H7 on cattle hides has been reported from 11% *(8)* to 76% *(3)*, whereas the prevalence of *Salmonella* on cattle hides has been as high as 94% *(7)*. Knowing the prevalence of these pathogens at different sites on beef cattle hides in the feedlot and at slaughter is essential for properly focusing on pathogen reducing interventions. However, the differences observed between studies have raised questions as to the best site on hides from which to collect samples. A better understanding of the distribution of these pathogens will aid in the determination of sampling locations that provide optimal detection for measuring pathogen prevalence. Previous studies have focused on the prevalence of these pathogens at various sites on cattle hides in the feedlot environment, and conflicting results have been reported *(12, 18)*. The prevalence of these pathogens is now becoming more important as treatments are focused on animals before entry to the food chain and on parts of the hide that can spread pathogens during the hide removal process in the plant. To our knowledge, no such studies have been performed at the processing plant, neglecting the effects of the lairage environment contamination events *(2)*.

In most studies, hide samples from only one side of the animal have been collected, which may lead to an underestimation of the true pathogen prevalence. For example, right-handed animals lie on one side predominantly. Therefore, the pathogens transferred from feedlot surfaces may not be evenly distributed across the sides *(19)*. The objective of this study was to reconcile differences among results from studies in which samples have been collected from different sites on the hide and to determine whether an optimal hide sampling site can be recommended. To this end, the prevalence of *E. coli* O157:H7 and *Salmonella* among multiple sites on beef cattle hides in two distinct production environments, a feedlot and a commercial processing plant, was determined and analyzed.

MATERIALS AND METHODS

Design. Two studies were conducted, one at a feedlot and the other at a commercial processing plant. Six sites were sampled on the hide of each animal at the feedlot to determine the prevalence rates of *E. coli* O157:H7 and *Salmonella*. Because of space constraints at the processing plant, only four of the hide sample

* Author for correspondence. Tel: 402-762-4224; Fax: 402-762-4149; E-mail: norasak.kalchayanand@ars.usda.gov.

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‡ Present address: IEH Laboratories & Consulting Group, 15300 Bothell Way N.E., Lake Forest Park, WA 98155, USA.

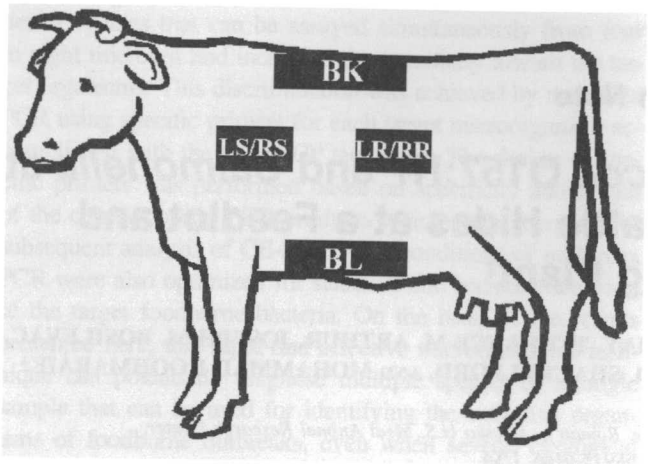

FIGURE 1. *Sampling sites of each animal at a feedlot. Approximately 1,000 cm² of each of the following areas were sampled: LS, left shoulder; RS, right shoulder; LR, left side of rib; RR, right side of rib; BK, back; BL, belly.*

sites on each animal were accessible. Samples were collected on four separate trips for both the feedlot and the processing plant.

Feedlot study. Hide samples were collected from 256 cattle at a 5,000-head feedlot at the U.S. Meat Animal Research Center (Clay Center, NE) from June to November. A sterile sponge (Nasco, Ft. Atkinson, WI) premoistened with 20 ml of buffered peptone water (Becton Dickinson, Sparks, MD) was used as a swab. Cattle were restrained in a squeeze chute, and approximately 1,000 cm^2 (an area approximately 30.3 by 33.0 cm) at each of the following sites on each animal were sampled: the left and the right shoulders (posterior half of shoulder to the fourth rib and midway between the back and the brisket; LS and RS), the left and the right side of ribs (12th or 13th rib area midway between the back and the belly; LR and RR), the back (dorsal thoracic midline; BK), and the belly (ventral abdominal midline; BL) (Fig. 1) using five strokes (one motion back and forth was considered a stroke) for each side of the sponge *(3)*. The sponges were placed in the bags and transported back to the laboratory for sample processing.

In-plant study. Hide samples were collected at a large central U.S. processing plant between March and September. Randomly selected carcasses ($n = 225$) were sampled on-line during processing immediately after stunning and exsanguination. Hide samples (1,000 cm^2) from each animal were collected with a sterile premoistened sponge (as described above) from four sites: LS, RS, BK, and BL. The sponge samples were placed in a cooler containing ice packs and transported back to the laboratory within 2 h for microbiological analyses.

Microbiological and statistical analyses. The sponge samples were processed for prevalence of *E. coli* O157:H7 and *Salmonella* in a nonselective enrichment culture according to the methods previously described, with slight modifications *(5, 15)*. One milliliter of each enrichment culture was first mixed with 20 μl of anti-*Salmonella* magnetic beads (Invitrogen, Carlsbad, CA) and subjected to immunomagnetic separation (IMS) as previously described *(14)*. The *Salmonella*-bead complexes (100 μl) were directly transferred into 3 ml of Rappaport-Vassiliadis-soya broth (RVS; Remel, Lenexa, KS) for selective enrichment and incubated at 42°C for 18 to 24 h. *Salmonella* present in these samples was detected by streaking for isolation of the RVS enrichment onto brilliant green agar with sulfadiazine (Becton Dickinson) and Hektoen enteric agar (Becton Dickinson) supplemented with 15 mg/liter novobiocin. The plates were incubated at 37°C for 18 to 20 h. Three presumptive colonies were confirmed as *Salmonella* based on the presence of *invA* gene by DNA PCR *(16)*.

Following *Salmonella* IMS, 20 μl of anti-O157 magnetic beads (Invitrogen) was added to the same 1-ml enrichment aliquots, and the bacteria-bead complexes were recovered as described for *Salmonella*. Fifty microliters of the *E. coli* O157–bead complexes was spread plated onto CHROMAgar O157 (DRG International, Mountainside, NJ) supplemented with 5 mg/liter novobiocin and 1.0 mg/liter potassium tellurite (Sigma, St. Louis, MO), and another 50 μl of complex was spread plated onto sorbitol MacConkey agar (Becton Dickinson) containing 0.05 mg/liter cefixime and 2.5 mg/liter potassium tellurite (Invitrogen). All plates were incubated at 37°C for 18 to 20 h, and up to three presumptive colonies were confirmed as harboring genes for the O157 antigen, H7 flagella, and at least one of the Shiga toxins using a multiplex PCR assay *(11)*.

For each sample site, prevalence of each pathogen was calculated by dividing the number of animals with a positive result by the total of number of animals sampled. To test for sample site prevalence differences for each pathogen, the DIFFER procedure of the PEPI software (USD, Inc., Stone Mountain, GA) *(9)* was used to calculate the pairwise differences among sites with significance defined at $P \leq 0.05$.

RESULTS AND DISCUSSION

Feedlot study. *Salmonella* prevalence was only 7% and therefore was not sufficient for the pathogen mapping analysis described here. The prevalence of *E. coli* O157:H7 did not differ ($P > 0.05$) among sampling trips 1, 3, and 4. Although trip 2 samples had a significantly lower prevalence of *E. coli* O157:H7 ($P \leq 0.05$), 91.0% of animals overall (data not shown) were positive for the pathogen at one or more sampling sites. The number of overall animals (97.8%) that were positive for *E. coli* O157:H7 at one or more hide locations across all sampling sites and trips were calculated (data not shown). These calculations revealed that that the hide is a major source of this pathogen, which may be transferred to carcasses during hide removal *(4, 6, 15)*. The rate of detection of *E. coli* O157:H7 on cattle hides across all sampling sites and sampling trips ranged from 17.8 to 100.0% (Table 1). In our study, sampling on both left and right sides was used to determine whether differences between hide sample sites existed. The belly was included because of its importance in hide opening and removal during slaughter. The back was included based on data collected by other researchers who suggested this area was the optimal location for *E. coli* O157:H7 detection *(12)*. The overall frequency of *E. coli* O157:H7 recovery among sampling sites ranged between 68.4 and 92.2%. There was no difference ($P \geq 0.05$) among four of the sampling sites (LS, RS, LR, and RR) in prevalence of *E. coli* O157:H7. However, the belly samples had the highest rate of recovery ($P \leq 0.05$) for *E. coli* O157:H7 (92.2%), and the back had the lowest recovery rate ($P \geq 0.05$) (68.4%). The finding of the lowest prevalence of *E. coli* O157:H7 on the back in this study is in contrast to previous reports that the highest prevalence of *E. coli* O157:H7 on cattle hides was the back *(12)*. These differences in the results might be attributed to the enrichment and detection methods. Keen and Elder *(12)* collected samples (500 cm^2)

TABLE 1. *Distribution of* E. coli *O157:H7 at different sites on cattle hides at a feedlot*[a]

Trip (month)	*n*	No. (%) of positive sites[b]					
		Left shoulder	Right shoulder	Left rib	Right rib	Back	Belly
1 (June)	50	46 (92.0) c	49 (98.0) c	49 (98.0) c	47 (94.0) c	40 (80.0) c	48 (96.0) c
2 (June)	56	14 (25.0) d	10 (17.8) d	14 (25.0) d	12 (21.4) d	12 (21.4) d	42 (75.0) d
3 (August)	100	98 (98.0) c	96 (96.0) c	95 (95.0) c	97 (97.0) c	78 (78.0) c	97 (97.0) c
4 (November)	50	50 (100.0) c	49 (98.0) c	49 (98.0) c	50 (100.0) c	45 (90.0) c	49 (98.0) c
Prevalence (%)		208 (81.2) A	204 (79.7) A	207 (80.8) A	206 (80.5) A	175 (68.4) B	236 (92.2) C

[a] Approximately 1,000 cm^2 was swabbed with sponges for each sample from each sampling site on each animal.
[b] Within a column, values with a common lowercase letter do not differ significantly ($P \geq 0.05$). Within the prevalence row, values with a common uppercase letter do not differ significantly ($P \geq 0.05$).

and used a selective medium for enrichment. Use of this selective enrichment medium may have resulted in an underestimation of *E. coli* O157:H7 prevalence because the environmentally stressed bacterial cells could have had difficulty proliferating on the selective medium *(17)*. Stephens et al. *(18)* used a nonselective enrichment method, which allowed stressed bacterial cells to recover before culture on selective medium, and found that the back samples had relatively low prevalence of *E. coli* O157:H7. These researchers reported that the samples with highest prevalence were taken at the hock and perineal areas *(18)*. However, these areas were avoided in the current study because they were considered to be more representative of the fecal shedding of the individual animal than of the other contamination sources present in the feedlot.

In the present study, samples were collected from both the left and right sides to determine whether the report of cattle "sidedness" *(19)* might lead to an underestimation of prevalence. Our results indicated no differences between left and right sides. However, the cattle sampled had not been studied for "handedness" behavior, so the predicted versus determined prevalence of *E. coli* O157:H7 is not known. The most heavily contaminated area of the hide was the belly area. This area of hide was included because it is part of a pattern line opened during hide removal and poses a risk for direct hide-to-carcass contamination. Our results indicated that for monitoring of *E. coli* O157:H7 in a feedlot environment, a sample collected from the belly area provides the most accurate indication of animal and pen prevalence.

Processing plant study. Because of the inaccessibility of the upper portions of the shackled cattle at the processing plant, hide samples could not be collected properly at the left and right rib and/or short rib areas. Thus, only four hide sites (LS, RS, BK, and BL) were sampled for each animal at the processing plant. A total of 225 animals were sampled to determine the prevalence of *E. coli* O157:H7 and *Salmonella* (Table 2). The rate of recovery for *E. coli* O157:H7 from all sampling sites and sampling trips ranged from 6.4 to 100.0% (Table 2). The overall frequency of detecting *E. coli* O157:H7 at any one sample site ranged from 76 to 78.7% across sampling sites. There was no difference ($P \geq 0.05$) in the prevalence of *E. coli* O157:H7 among the LS, RS, BK, and BL sites. This apparent equal distribution of the pathogen across sampling sites is consistent with additional hide contamination that occurs during transportation to and lairage at the processing plant *(2, 13)*. The first trip to the processing plant was during winter, and samples collected at this time had the lowest prevalence ($P \leq 0.05$) of *E. coli* O157:H7. Barkocy-Gallagher et al. *(4)* reported that the prevalence of *E. coli* O157:H7 on hide

TABLE 2. *Distribution of* E. coli *O157:H7 and* Salmonella *at different sites on cattle hides at a processing plant*[a]

Organism	Trip (month)	*n*	No. (%) of positive sites[b]			
			Left shoulder	Right shoulder	Back	Belly
E. coli O157:H7	1 (March)	47	3 (6.4) e	4 (8.5) e	3 (6.4) d	3 (6.4) d
	2 (June)	64	54 (84.4) c	57 (89.1) c	60 (93.8) c	61 (95.3) c
	3 (August)	64	64 (100.0) d	64 (100.0) d	64 (100.0) c	64 (100.0) c
	4 (September)	50	50 (100.0) d	49 (98.0) cd	49 (98.0) c	49 (98.0) c
	Prevalence (%)		171 (76.0) A	174 (77.3) A	176 (78.2) A	177 (78.7) A
Salmonella	1 (March)	47	42 (89.4) d	44 (93.6) e	42 (89.4) e	46 (97.9) d
	2 (June)	64	17 (26.6) c	13 (20.3) c	6 (9.4) c	35 (54.7) c
	3 (August)	64	14 (21.9) c	17 (26.6) c	17 (26.6) c	33 (51.6) c
	4 (September)	50	37 (74.0) d	34 (68.0) d	24 (60.0) d	38 (76.0) d
	Prevalence (%)		110 (48.9) A	108 (48.0) A	89 (39.5) B	152 (68.2) C

[a] Approximately 1,000 cm^2 was swabbed with sponges for each sample from each sampling site on each animal.
[b] For each organism, within a column, values with a common lowercase letter do not differ significantly ($P \geq 0.05$). Within each prevalence row, values with a common uppercase letter do not differ significantly ($P \geq 0.05$).

TABLE 3. Salmonella *prevalence for two combined sampling sites on cattle hides at a processing plant*

Pooled trip (month)	*n*	No. (%) of positive combined sites[a]					
		LS and BL	RS and BL	BK and BL	LS and RS	LS and BK	RS and BK
1 (March)	47	47 (100.0) d	47 (100.0) e	47 (100.0) d	47 (100.0) d	46 (97.9) d	46 (97.9) d
2 (June)	64	39 (60.9) c	37 (57.8) c	36 (56.2) ce	24 (37.5) c	20 (31.2) c	17 (26.6) c
3 (August)	64	40 (62.5) c	40 (62.5) d	40 (62.5) e	28 (43.8) c	27 (42.2) c	31 (48.4) e
4 (September)	50	43 (86.0) d	41 (82.0) de	42 (84.0) d	42 (84.0) d	43 (86.0) d	37 (74.0) d
Prevalence (%)		169 (75.1) A	165 (73.3) A	165 (73.3) A	141 (62.7) B	136 (60.4) B	131 (58.7) B

[a] LS, left shoulder; BL, belly; RS, right shoulder; BK, back. Within a column, values with a common lowercase letter do not differ significantly ($P \geq 0.05$). Within the prevalence row, values with a common uppercase letter do not differ significantly ($P \geq 0.05$).

differed by season, and pathogen recovery was lowest in winter.

For *Salmonella,* the prevalence across all sampling sites and sampling trips ranged from 9.4 to 97.9% (Table 2). The LS site did not differ ($P \geq 0.05$) from the RS sample in *Salmonella* detection, but both the LS and RS samples had higher prevalence ($P \leq 0.05$) than the BK samples. *Salmonella* was more often recovered ($P \leq 0.05$) from the BL samples (68.2%), whereas the BK was the site with the lowest *Salmonella* detection rate (39.5%).

Because the *Salmonella* prevalence was significantly different between some of the sites, these data were examined in greater detail. *Salmonella* prevalence data for each site from each trip to the processing plant were analyzed by pooling the six two-way combinations of LS, RS, BK, and BL to evaluate whether any two combined sampling sites would improve the rate of *Salmonella* detection (Table 3). Overall, the combination of LS and RS (62.7%), LS and BK (60.4%), or RS and BK (58.7%) had lower prevalences of *Salmonella* at the plant when compared with any sample that included BL. Sampling combinations of LS and BL (75.1%), RS and BL (73.3%), or BK and BL (73.3%) were not different ($P \geq 0.05$) from each other. However, these three combined sites resulted in a higher frequency of *Salmonella* detection at the plant than did sampling only the BL (68.2%; Table 2). Multiple sampling sites also have been suggested to more accurately reflect the prevalence of *E. coli* O157:H7 and *Salmonella (12, 18).*

Our study involved mapping the natural contamination of *E. coli* O157:H7 and *Salmonella* on cattle hides at a feedlot and a processing plant. This mapping can provide useful information to the processor concerning which sites on incoming animals should be the focus of interventions and which sample sites can provide the most representative prevalence data for various pathogens. The belly along the ventral abdominal midline was most likely to yield *E. coli* O157:H7 at the feedlot and *Salmonella* at the processing plant. Sampling at two sites, such as the belly with one side of the sponge and the left, right shoulder, or back along a dorsal thoracic midline with the other side of the sponge, provided a higher rate of *Salmonella* detection than did sampling the belly alone. Our results identified the belly as an essential target for decontamination to prevent hide-to-carcass contamination because this area is part of the pattern line to be opened during hide removal.

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REFERENCES

1. Anonymous. 2006. Foodborne illness cost calculator. U.S. Department of Agriculture, Economic Research Service. Available at: http://www.ers.usda.gov/Data/FoodborneIllness. Accessed 28 January 2009.
2. Arthur, T. M., J. M. Bosilevac, D. M. Brichta-Harhay, M. N. Guerini, N. Kalchayanand, S. D. Shackelford, T. L. Wheeler, and M. Koohmaraie. 2007. Transportation and lairage environment effects on prevalence, numbers, and diversity of *Escherichia coli* O157:H7 on hides and carcasses on beef cattle at processing. *J. Food Prot.* 70: 280–286.
3. Arthur, T. M., J. M. Bosilevac, X. Nou, S. D. Shackelford, T. L. Wheeler, M. P. Kent, D. Jaroni, B. Pauling, D. M. Allen, and M. Koohmaraie. 2004. *Escherichia coli* O157:H7 prevalence and enumeration of aerobic bacteria, *Enterobacteriaceae,* and *Escherichia coli* O157:H7 at various steps in commercial beef processing plants. *J. Food Prot.* 67:658–665.
4. Barkocy-Gallagher, G. A., T. M. Arthur, M. Rivera-Betancourt, X. Nou, S. D. Shackelford, T. L. Wheeler, and M. Koohmaraie. 2003. Seasonal prevalence of Shiga toxin–producing *Escherichia coli* O157:H7 and non-O157:H7 serotypes and *Salmonella* in commercial beef processing plants. *J. Food Prot.* 66:1978–1986.
5. Barkocy-Gallagher, G. A., K. K. Edwards, X. Nou, J. M. Bosilevac, T. M. Arthur, S. D. Shackelford, and M. Koohmaraie. 2005. Methods for recovering *Escherichia coli* O157:H7 from cattle fecal, hide, and carcass samples: sensitivity and improvements. *J. Food Prot.* 68: 2264–2268.
6. Bosilevac, J. M., T. M. Arthur, T. L. Wheeler, S. D. Shackelford, M. Rossman, J. O. Reagan, and M. Koohmaraie. 2004. Prevalence of *Escherichia coli* O157 and levels of aerobic bacteria and *Enterobacteriaceae* are reduced when hides are washed and treated with cetylpyridinium chloride at a commercial beef processing plant. *J. Food Prot.* 67:646–650.
7. Brichta-Harhay, D. M., M. N. Guerini, T. M. Arthur, J. M. Bosilevac, N. Kalchayanand, S. D. Shackelford, T. L. Wheeler, and M. Koohmaraie. 2008. *Salmonella* and *E. coli* O157:H7 contamination on hides and carcasses of cull cattle presented for slaughter in the United States—an evaluation of prevalence and load using immunomagnetic separation and direct plating methods. *Appl. Environ. Microbiol.* 74:6289–6297.
8. Elder, R. O., J. E. Keen, G. R. Siragusa, G. A. Barkocy-Gallagher, M. Koohmaraie, and W. W. Laegreid. 2000. Correlation of enterohemorrhagic *Escherichia coli* O157:H7 prevalence in feces, hides, and carcasses of beef cattle during processing. *Proc. Natl. Acad. Sci. USA* 97:2999–3003.

9. Fliess, J. L. (ed.). 1981. Statistical methods for rates and proportions, 2nd ed. John Wiley and Sons, New York.
10. Grandin, T. 1990. Handling practices in U.S. feedlots and packing plants, p. 115–120. *In* Proceedings of the Livestock Conservation Institute. Livestock Conservation Institute, Madison, WI.
11. Hu, Y., Q. Zhang, and J. C. Meitzler. 1999. Rapid and sensitive detection of *Escherichia coli* O157:H7 in bovine faeces by a multiplex PCR. *J. Appl. Microbiol.* 87:867–876.
12. Keen, J. M., and R. O. Elder. 2002. Isolation of Shiga-toxigenic *Escherichia coli* O157:H7 from hide surfaces and the oral cavity of finishing beef feedlot cattle. *J. Am. Vet. Med. Assoc.* 220:756–763.
13. Matthew, L., J. C. Low, D. L. Gally, M. C. Pearce, D. J. Mellor, J. A. Heesterbeck, M. Chase-Topping, S. W. Naylor, D. J. Shaw, S. W. Reid, G. J. Gunn, and M. E. Woolhouse. 2006. Heterogeneous shedding of *Escherichia coli* O157:H7 in cattle and its implications for control. *Proc. Natl. Acad. Sci. USA* 103:547–552.
14. Nou, X., T. M. Arthur, J. M. Bosilevac, D. M. Brichta-Harhay, M. N. Guerini, N. Kalchayanand, and M. Koohmaraie. 2006. Improvement of immunomagnetic separation for *Escherichia coli* O157:H7 detection by the PickPen magnetic particle separation device. *J. Food Prot.* 69:2870–2874.
15. Nou, X., M. Rivera-Betancourt, J. M. Bosilevac, T. L. Wheeler, S. D. Shackelford, B. L. Gwartney, J. O. Reagan, and M. Koohmaraie. 2003. Effect of chemical dehairing on the prevalence of *Escherichia coli* O157:H7 and the levels of aerobic bacteria and *Enterobacteriaceae* on carcasses in a commercial beef processing plant. *J. Food Prot.* 66:2005–2009.
16. Rahn, K., S. A. DeGrandis, R. C. Clarke, S. A. McEwen, J. E. Galan, C. Ginocchio, R. Curtiss, and C. L. Gyles. 1992. Amplification of an *invA* gene sequence of *Salmonella typhimurium* by polymerase chain reaction as a specific method of detection of *Salmonella. Mol. Cell. Probes* 6:271–279.
17. Ray, B. 2001. Microbial sporulation and sublethal injury, p. 79–94. *In* B. Ray (ed.), Fundamental food microbiology. CRC Press, Boca Raton, FL.
18. Stephens, T. P., G. H. Loneragan, T. W. Thompson, A. Sridhara, L. A. Branham, S. Pitchiah, and M. H. Brashears. 2007. Distribution of *Escherichia coli* O157:H7 and *Salmonella* on hide surfaces, the oral cavity, and in feces of feedlot cattle. *J. Food Prot.* 70:1346–1349.
19. Wagnon, K. A., and W. C. Rollins. 1972. Bovine laterality. *J. Anim. Sci.* 35:486–488.