Translocation of Surface-Inoculated *Escherichia coli* O157:H7 into Beef Subprimals following Blade Tenderization†‡

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ABSTRACT

In phase I, beef subprimals were inoculated on the lean side with ca. 0.5 to 3.5 log CFU/g of a rifampin-resistant (rifr) cocktail of *Escherichia coli* O157:H7 and passed once, lean side up, through a mechanical blade tenderizer. Inoculated subprimals that were not tenderized served as controls. Ten core samples were removed from each sub primal and cut into six consecutive segments: segments 1 to 4 comprised the top 4 cm and segments 5 and 6 the deepest 4 cm. Levels of *E. coli* O157:H7 recovered from segment 1 of control subprimals when inoculated with ca. 0.5, 1.5, 2.5, or 3.5 log CFU/g were 0.6, 1.46, 2.5, and 3.19 log CFU/g, respectively. Following tenderization, pathogen levels recovered from segment 1 inoculated with 0.5 to 3.5 log CFU/g were 0.22, 1.06, 2.04, and 2.7 log CFU/g, respectively. Levels recovered in segment 2 were 7- to 34-fold lower than levels recovered from segment 1. Next, in phase II, the translocation of ca. 4 log CFU of the pathogen per g was assessed for lean-side–inoculated subprimals passed either once (LS) or twice (LD) through the tenderizer and for fat-side–inoculated subprimals passed either once (FS) or twice (FD) through the tenderizer. Levels in segment 1 for LS, LD, FS, and FD tenderized subprimals were 3.63, 3.52, 2.85, and 2.55 log CFU/g, respectively. The levels recovered in segment 2 were 7- to 50-fold lower than levels recovered in segment 1 for LS, LD, FS, and FD subprimals. Thus, blade tenderization transfers *E. coli* O157:H7 primarily into the topmost 1 cm, but also into the deeper tissues of beef subprimals.

Illnesses due to *Escherichia coli* O157:H7 have been linked to a variety of sources and foods, but undercooked ground beef is arguably the primary vehicle (6). It is noteworthy that outbreaks have been linked occasionally to intact (nonground) beef and that relatively recent outbreaks have also been presumably associated with nonintact, that is, mechanically tenderized, steaks (1, 2, 15, 20). Over a 5-week period during June and July of 2002 the prevalence of *E. coli* O157:H7 on intact sub primal beef cuts just prior to mechanical tenderization from six beef processing plants across the United States was estimated at 0.2% (2 positive samples among 1,014 samples tested) using a PCR-BAX method (2, 21). Additionally, for the two positive samples, pathogen levels were estimated at <0.375 CFU/cm² using a most-probable-number method (21). Other investigators reported the incidence of *E. coli* O157:H7 on beef subprimals at <0.083% from among nearly 1,200 samples tested (2). Perhaps more importantly, from a food safety perspective, bacteria are transferred from the surface into the deeper tissues of subprimals following mechanical tenderization. For example, Gill et al. (5) reported that aerobes, coliforms, and staphylococci/listeriae, but not *E. coli*, were recovered from the deep tissues of meat tenderized at a packing plant. In addition, these same types of bacteria were recovered from the deep tissues of meat that were mechanically tenderized at retail (4). As another example, Sporing (14) reported that single-pass, blade-tenderization processing of surface-contaminated, intact beef muscle (inside rounds) internalized approximately 3 to 4% of the initial inoculum of *E. coli* O157:H7 into the geometric center of the steaks subsequently obtained from these inside rounds. In related studies using *Salmonella* inoculated onto pork, Lambert et al. (10) also reported that blade tenderization internalized 1 to 7% of the cells from the surface into the deeper tissues of the muscle, and that needle injection transferred 4 to 8% from the surface to the center of the cut. The potential for contamination and subsequent transfer of pathogens such as *E. coli* O157:H7 into the interior of mechanically tenderized meat and the potential for infections if such products are not adequately cooked resulted, in part, in the proposed intention of the U.S. Department of Agriculture/Food Safety and Inspection Service (USDA/FSIS) to expand the *E. coli* O157:H7 adulteration policy to include nonintact products (16, 17). This amended policy now requires manufacturers to reassess their hazard analysis and critical control point (HACCP) plans for mechanically tenderized products to address relevant biological hazards, particularly those presented by *E. coli* O157:H7 (20). Beef products possibly affected would include primal and subprimal cuts that are blade tenderized or needle injected. Since these cuts will probably be perceived by both consumers and retail stab-
lishments as “like” intact steaks/roasts and as “not like”
ground beef, on occasion, these cuts may not be cooked to
internal temperatures that would destroy any surface mi-
crobes transferred into the interior of these tenderized or
restructured products. Thus, the objective of this study was
to assess the extent of penetration and/or transfer of differ-
et levels of *E. coli* O157:H7 inoculated onto either the fat-
or the lean-side surfaces of top cut subprimals that were passed either once or twice through a mechanical blade tenderizer.

### MATERIALS AND METHODS

**Bacterial strains.** Approximately equal numbers of each of the following five rifampin-resistant (rifR) strains of *E. coli* O157:
H7 reported previously by Phebus et al. (13) and Sporing (14) were used as a cocktail in this study: (i) USDA-011-82 (meat outbreak); (ii) ATCC 43888 (human isolate, CDC B6914-MS-1); (iii) ATCC 43889 (human isolate, CDC B1409-C1); (iv) ATCC 43890 (human isolate, CDC C984); and (v) USDA-FSIS 45756 (meat isolate). It should be noted that Sporing (14) used only
strain USDA-011-82 for the translocation component of their study and all five of the above-mentioned strains for the cooking component. They generated rifR derivatives of these strains by se-
quential transfer on agar plates containing increasing levels of
rifampin as described earlier (14). To prepare the cocktail, a loop-
ful of an isolated colony of each of the five strains was transferred
to separate test tubes containing 10 ml of tryptic soy broth (Difco
Laboratories Inc., Detroit, Mich.) and incubated for 18 to 20 h at
37°C without shaking. The entire 10 ml of each of the freshly
grown cultures was combined in a 50-ml sterile conical tube and
centrifuged at 5,000 × g for 10 min at 4°C. Cell pellets were
resuspended in 50 ml of sterile 0.1% peptone water (PW; Difco),
serially diluted in PW to reach the target inoculum level, and held
at 4°C for about 30 min.

**Inoculation and mechanical tenderization of subprimals.**
Top cut beef subprimals (ca. 15 to 20 lb each) were purchased
from a local wholesale distributor and stored at 4°C for up to 7
days. A total of 64 top cut subprimals were obtained over eight
separate visits to the distributor from December 2004 through No-

These experiments were conducted in the Pathogen Compatible
Food Processing Suite at the USDA/Agricultural Research Ser-
vice/Eastern Regional Research Center (Wyndmoor, Pa.) using
equipment and resources dedicated specifically for use in this fa-
cility. Each of the two blade sets consisted of seven rows that
each contained 12 stainless steel blades (3 mm wide each), with
alternate blades being set with the width parallel or at right angles
to the direction of travel of the conveyor belt. Each set of blades
descended until the tips were within 2 mm of the conveyor belt.
The conveyor belt advanced by increments of 3.6 cm and the two
sets of blades descended and penetrated the subprimal after each
advancement of the belt. Thus, each subprimal was tenderized by
both sets of blades. After each trial, the blade sets were removed
from the tenderizer and thoroughly sprayed with hot (190°F) water
to remove debris and then were soaked for up to 2 h in a 1:256
solution of quaternary ammonium-based detergent (BDD, Koch
Supplies, Kansas City, Mo.). The tenderizer itself, including the
conveyor belt and the housing for the blade set, was also sprayed
with hot water and then wiped down with the same detergent.

**Phase I. Effect of inoculation level on translocation fol-
lowing lean-side inoculation and single-pass tenderization.**
Each subprimal was aseptically removed from its original pack-
aging and placed into a sterile plastic bag with the lean side of
the subprimal facing upward. Ten milliliters of the bacterial cock-
tail was applied to the lean side by using a pipette to achieve a
target level of about 0.5, 1.5, 2.5, or 3.5 log CFU/g. As an aside,
this corresponds to about 0.5, 1.5, 2.5, or 3.5 log CFU/cm² of
the surface area available for inoculating the lean (or fat) side of
subprimals. The opening of each bag was then sealed with tape, and
the inoculated subprimals were stored, with the inoculated surface
facing down, for ca. 30 min at 4°C to allow the weight of the
subprimal to distribute the inoculum over the surface and to pro-
mote attachment of the cells to the meat. Next, to aid in transfer
and handling, each subprimal was placed onto polystyrene foam
packing trays (Koch Supplies) so that the inoculated side was
facing up and passed through the tenderizer. One set of lean-side–
inoculated subprimals was passed longitudinally through the
tenderizer once (LS), and another set of the inoculated subprimals
was not tenderized and served as the positive controls. A total of
two trials were conducted for each inoculation level, with a single
trial consisting of two tenderized subprimals and two positive-
control, i.e., nontenderized, subprimals. A total of 10 core samples
(Fig. 2A) were obtained from each of the subprimals for each trial
and for each inoculation level: 320 core samples (1 treatment [LS]
× 4 inoculation levels × 2 experimental subprimals per treatment
× 2 control subprimals per treatment × 2 trials per inoculum level

**Phase II. Effect of blade tenderization.** The tenderizer was
run for 2 min at the speed designated for beef. The rest of the
protocol was the same as described above in Phase I.
× 10 core samples per subprimal) were obtained from 32 subprimals and sampled for E. coli O157:H7.

Phase II. Effect of fat- versus lean-side and single- versus double-pass tenderization on translocation. As described in the preceding section, subprimals were inoculated with 10 ml of the five-strain cocktail of E. coli O157:H7 to achieve a target level of about 4 log CFU/g. One set of subprimals, with the inoculated fat or lean side facing up, were passed longitudinally through the tenderizer once. Another set of the inoculated subprimals were not tenderized and served as the positive controls. Double-pass tenderized subprimals were passed once through the tenderizer, rotated 180° horizontally, and then passed a second time in the opposite direction relative to the first pass. For this study, subprimals were subjected to a total of four tenderization treatments as follows: (i) lean-side inoculation, single-pass tenderization (LS); (ii) lean-side inoculation, double-pass tenderization (LD); (iii) fat-side inoculation, single-pass tenderization (FS); and (iv) fat-side inoculation, double-pass tenderization (FD). A total of two trials were conducted for each treatment, with a single trial consisting of two tenderized subprimals and two positive-control, i.e., non-tenderized, subprimals. A total of 10 core samples were obtained from each of the subprimals for each trial and for each treatment for a total of 320 core samples (4 treatments [LS, LD, FS, and FD] × 1 inoculation level × 2 experimental subprimals per treatment × 2 control subprimals per treatment × 2 trials per treatment × 10 core samples per subprimal) from 32 subprimals.

Microbiological sampling of core samples. After tenderization, subprimals were aseptically transferred to an alcohol-sterilized polystyrene cutting board (24 by 36 in.) with the inoculated surface facing down so that the core samples likely to contain the fewest cells of E. coli O157:H7 could be extracted from the surface. Ten core samples were obtained from each tenderized subprimal by using a sterile, stainless steel fabricated metal coring device (Fig. 2B; ca. 10 cm in length and ca. 5 cm in diameter) inserted into the meat from the noninoculated surface. The resulting core sample was aseptically removed from the coring device and placed horizontally onto clean butcher paper. To minimize contamination, each core was carefully removed from the coring device starting from the noninoculated surface, that being the bottom of the subprimal, to prevent the sample from passing through the inoculated surface end of the coring device. Core samples were then subsequently cut into six consecutive segments by using an alcohol-sterilized knife starting from the inoculated surface: segments 1 through 4 comprised the top 4 cm of the core, and segments 5 and 6 comprised the lowest 4 cm of the core (Fig. 3). The knife was sterilized with alcohol between each cut. Ten core samples were also obtained from approximately the same locations on the surface of positive-control subprimals by using the same coring device as that described above. Each segment was weighed using a top-loading balance (model SP202, Ohaus, Pine Brook, N.J.) to determine the weight of the meat and mash present in each segment. Composite samples (3) were collected in a sterile 50-ml plastic test tube. The rinse fluid was diluted as appropriate in PW and spread plated onto SMACR agar plates in the same manner described above.

Tenderized Surface

Lean Side

Fat Side

FIGURE 3. Photograph of core samples showing the approximate location of each of the six segments that were obtained. Segments 1 to 4 were 1 cm thick, and segments 5 and 6 were 2 cm each.

Statistical analyses. The transfer of E. coli O157:H7 cells into deeper tissues by way of mechanical tenderization was expressed (as a percentage) as the number of cells (CFU per gram) recovered separately from each of the six segments obtained from tenderized subprimal cores, divided separately by the number of cells (CFU per gram) recovered from segment 1 of the cores obtained from the nontenderized, positive-control subprimals. The standard deviations for the levels of the pathogen recovered from each of the six segments and the cumulative totals recovered from core samples were calculated using the statistical function option that is provided with Microsoft Excel 2003 software (Redmond, Wash.). Data were analyzed using version 8.0 of the SAS statistical package (SAS Institute, Inc., Cary, N.C.). Analyses were performed using an unstructured variance-covariance model to evaluate the effects and interactions of each inoculation level for LS, LD, FS, and FD subprimals on the levels of E. coli O157:H7. Additionally, a mixed-model procedure of the SAS software was used to analyze the effects and interactions of inoculation level, following the USDA/FSIS E. coli O157:H7 isolation/enrichment protocol (3). For LS subprimals that were inoculated with four levels of E. coli O157:H7, segments 1 and 2 from each core sample that tested negative for the pathogen by direct plating were enriched separately. However, due to the high number of samples that tested negative by direct plating, it was necessary to combine segments 3 through 6 for a core sample for each subprimal to test for the presence or absence of the pathogen by enrichment. Briefly, 10 ml of the blended or macerated fluid was transferred from each segment 3 through segment 6 composite sample and combined in a sterile plastic vacuum bag and processed following the USDA/FSIS enrichment protocol (3). In addition, at the conclusion of each trial, the blades from the tenderizer were also tested for the presence and levels of E. coli O157:H7. Briefly, a sterile sponge (Nasco, Ft. Atkinson, Wis.) was rehydrated with 25 ml of sterile PW and used to swab the outer surface of the outermost set of blades following the tenderization of two subprimals. After swabbing, each sponge was agitated in a stomacher for 2 min to loosen cells from the sponge, and the resulting rinse fluid was collected in a sterile 50-ml plastic test tube. The rinse fluid was diluted as appropriate in PW and spread plated onto SMACR agar plates in the same manner described above.
tenderization method (single versus double pass), and meat surface (lean- versus fat-side inoculation and tenderization) on the levels of the pathogen that were recovered from each of the six segments from each of the 10 core samples for a given subprimal. Differences were considered significant at $P$ values of $\leq 0.05$.

**RESULTS**

**Phase I. Effect of inoculation level on transfer of** *E. coli* O157:H7 **into the lean side of subprimals passed once through the tenderizer.** Preliminary studies showed that more cells were transferred into the deeper tissues of lean-side–inoculated, single-pass subprimals (LS) than into lean-side–inoculated, double-pass subprimals (LD), or fat-side–inoculated, single-pass subprimals (FS), or fat-side–inoculated, double-pass subprimals (FD) (data not shown). Therefore, the effect of LS on the transfer of varying surface levels (ca. 0.5 to 3.5 log CFU/g) of *E. coli* O157:H7 into beef subprimals was investigated. The levels of *E. coli* O157:H7 transferred into all six segments of core samples ranged from 37 to 55%. Portions of the remaining 45 to 63% of the cells were found on the blades of the tenderizer (see below). It is also likely that some of the remaining cells would be found on the conveyor belt and other food

**TABLE 1. Levels of *E. coli* O157:H7 recovered from segmented core samples obtained from subprimals inoculated with different levels of the pathogen on the lean side and single-pass tendonization.**

<table>
<thead>
<tr>
<th>Inoculation level (log CFU/g)</th>
<th>% Recovery</th>
<th>% Transfer</th>
<th>Total level of <em>E. coli</em> O157:H7 transferred into all 6 segments of a core sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.45 (2.3)</td>
<td>40</td>
<td>1.9–2.76</td>
<td>$1.06 (1.20)$</td>
</tr>
<tr>
<td>1.9–2.76</td>
<td>2.77</td>
<td>1.0–1.20</td>
<td>$&lt;1.9–2.76$</td>
</tr>
<tr>
<td>1.9–1.20</td>
<td>1.38</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.20$</td>
</tr>
<tr>
<td>1.9–0.78</td>
<td>0.52</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–0.78$</td>
</tr>
<tr>
<td>1.9–0.78</td>
<td>0.26</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–0.78$</td>
</tr>
<tr>
<td>1.9–0.30</td>
<td>0.14</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–0.30$</td>
</tr>
<tr>
<td>1.9–0.60</td>
<td>0.33</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–0.60$</td>
</tr>
<tr>
<td>1.9–1.33</td>
<td>0.33</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.33$</td>
</tr>
<tr>
<td>1.9–1.96</td>
<td>0.16</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.96$</td>
</tr>
<tr>
<td>1.9–1.68</td>
<td>0.26</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.68$</td>
</tr>
<tr>
<td>1.9–1.96</td>
<td>0.52</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.96$</td>
</tr>
<tr>
<td>1.9–1.96</td>
<td>0.26</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.96$</td>
</tr>
<tr>
<td>1.9–1.68</td>
<td>0.16</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.68$</td>
</tr>
<tr>
<td>1.9–1.33</td>
<td>0.33</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.33$</td>
</tr>
<tr>
<td>1.9–1.00</td>
<td>0.14</td>
<td>0.0 (0)</td>
<td>$&lt;1.9–1.00$</td>
</tr>
</tbody>
</table>

- Percent transfer is calculated as (CFU per gram of tenderized core segment/CFU per gram of segment 1 of nontenderized control subprimal core) × 100.
- Range of *E. coli* (log CFU/g) transferred among 40 subprimals from 6 segments of a core sample.
contact surfaces, as well as in any purge, but these items were not sampled.

In total, 40 core samples (composed of segments 1 through 6) were obtained for each of the four inoculation levels of *E. coli* O157:H7 (2 experimental subprimals per inoculation level × 10 core samples per subprimal × 2 trials). Table 2 reports the number of core samples testing positive for the pathogen by direct plating, by enrichment, and by direct plating and enrichment combined for all six segments. Depending on the inoculation level, *E. coli* O157:H7 cells were recovered from segment 1 by direct plating from 50 to 98% (20 to 39) of each set of 40 core samples tested. However, for segment 2, the pathogen was recovered from 2.5 to 50% (1 to 20) of the 40 core samples depending on the inoculation level. When the results of direct plating and enrichment were combined, the pathogen was recovered from 90 to 100% (36 to 40) of the 40 core samples obtained from segment 1 and from 55 to 98% (22 to 39) of segment 2 core samples. Regardless of the inoculation level, relatively few (0 to 5) of the segment 3, 4, 5, or 6 samples tested positive for the pathogen by direct plating (Table 2). When samples from segments 3 through 6 were tested negative by direct plating for a given subprimal, segments 3 through 6 were combined and collectively enriched. By this approach, the pathogen was recovered from 38 to 70% of core samples comprised of segments 3 through 6 for each inoculation level. Based on the location of the 10 core samples and the levels of pathogen that were transferred into each core sample, there was no significant difference (*P* > 0.05) with regards to the level of the pathogen that was transferred into each of the six segments relative to cores 1 through 10 obtained from a given subprimal regardless of the inoculation level.

To determine if any cells of *E. coli* O157:H7 became associated with the tenderizer, the blade sets were sampled following tenderization of the two experimental subprimals for each trial. The pathogen was recovered at levels of 2.8, 3.2, 3.4, and 4.6 log CFU per two blade sets used to tenderize subprimals inoculated with 0.6, 1.46, 2.45, and 3.19 log CFU/g, respectively. Although it was not possible to directly compare the levels of cells found on the blades of the tenderizer to those inoculated onto the surface of the meat, as expected, these results demonstrate that the pathogen was found on the blades of the tenderizer. Additionally, because the blade sets penetrate through the entire subprimal, it is possible that some cells of the pathogen may have been transferred onto the conveyor belt and/or onto other parts of the machine in contact with the subprimal or blades during the tenderization process. These results may also explain, at least in part, why the total levels of *E. coli* O157:H7 recovered from the six segments of each core sample did not account for 100% of the initial inoculum levels that were applied to the surface of the subprimal. These findings also stress the importance of using an effective cleaning and sanitation protocol for the tenderizer and its component parts, but especially for the blades.

### Table 2. Direct plating and enrichment results for subprimals inoculated on the lean side with different levels of *E. coli* and then single-pass tenderized

<table>
<thead>
<tr>
<th>Segment no.</th>
<th>Initial levels (log CFU/g) of <em>E. coli</em> recovered from segment 1 of nontenderized subprimals (control):</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>3.19</td>
</tr>
<tr>
<td></td>
<td>Direct plating</td>
</tr>
<tr>
<td>1</td>
<td>38/40</td>
</tr>
<tr>
<td>2</td>
<td>20/40</td>
</tr>
<tr>
<td>3</td>
<td>5/40</td>
</tr>
<tr>
<td>4</td>
<td>2/40</td>
</tr>
<tr>
<td>5</td>
<td>5/40</td>
</tr>
<tr>
<td>6</td>
<td>4/40</td>
</tr>
</tbody>
</table>

- a Number of segments from which *E. coli* was recovered by direct plating from among 40 total segments from 40 core samples.
- b Number of segments from which *E. coli* was recovered by enrichment from segments negative by direct plating.
- c Composite sample containing segments 3 through 6 that were negative by direct plating and were enriched as a single sample (40 enrichments).
- d Number of segments from which *E. coli* was recovered by both direct plating and enrichment/total sampled (40 total).
than from segment 5. For each of the four treatments, there were significantly ($P < 0.05$) more cells of \textit{E. coli} O157:H7 transferred into segment 1 of the core samples than into segments 2 through 6 (data not shown). Pathogen numbers transferred into all six segments of core samples obtained from LS, LD, FS, and FD subprimals were 3.66, 3.56, 2.94, and 3.58 log CFU/g, respectively. In addition to the core samples, \textit{E. coli} O157:H7 cells were also recovered from the two blade sets for each of the two trials for LS, LD, FS, and FD subprimals at 4.6, 3.9, 3.9, and 3.0 log CFU per blade sets, respectively.

The total numbers of core samples from all six segments (40 total core samples from each segment for each treatment) from LS, LD, FS, and FD subprimals that tested positive for the pathogen by direct plating and/or by enrichment are provided in Table 4. When the results of both direct plating and enrichment sampling were taken into consideration, for all segments and all treatments the pathogen was recovered from 60 to 100% of all core samples tested. In addition, there were no statistical differences ($P > 0.05$) relative to the levels of the pathogen that were transferred into each of the six segments based on the location of the 10 core samples on the surface of the subprimal regardless of whether it was fat- or lean-side inoculated or regardless of the number of passes through the tenderizer (data not shown).

**DISCUSSION**

As evidenced by several large recalls of ground, non-intact beef between September 2007 (331,582 lb) and February 2008 (143 million pounds) (USDA/FSIS Recall Release; http://www.fsis.usda.gov/fsis.recalls/RecallCase_Archive/index.asp), \textit{E. coli} O157:H7 remains a significant public health concern and an appreciable economic burden to the food industry and consumers alike. In addition to recalls, there have also been illnesses related to nonintact beef that have sparked a renewed interest in determining the relative risk of such products to public health. More specifically, there was an outbreak in August 2000 linked with tenderized steaks from a restaurant, a second outbreak in June 2003 linked with a bacon-wrapped boneless beef with tenderized steaks from a restaurant, a second outbreak specifically, there was an outbreak in August 2000 linked with tenderized, marinated beef in February 2008 (143 million pounds) (USDA/FSIS Recall Release; http://www.fsis.usda.gov/fsis.recalls/RecallCase_Archive/index.asp), \textit{E. coli} O157:H7 remains a significant public health concern and an appreciable economic burden to the food industry and consumers alike. In addition to recalls, there have also been illnesses related to nonintact beef that have sparked a renewed interest in determining the relative risk of such products to public health. More specifically, there was an outbreak in August 2000 linked with tenderized steaks from a restaurant, a second outbreak in June 2003 linked with a bacon-wrapped boneless beef filet product injected with marinade, and a third outbreak in August 2004 linked with tenderized, marinated beef steak from a national restaurant chain (1, 2, 15, 20). In early 2007, there was an additional outbreak wherein several people became ill after consuming various steak products tenderized and prepared at a Pennsylvania restaurant (http://www.fsis.usda.gov/PDF/Recall_019_2007_Release.pdf). As part of an illness investigation there was also a recall of beef products, including mechanically tenderized steaks, in the spring of 2007 in Michigan (http://www.fsis.usda.gov/PDF/Recall_023_2007_Release.pdf). Such events emphasize the need for further discussion and possibly more research related to whether or not nonintact meat poses a greater threat to public health than otherwise similar meat that is not tenderized or injected.

In a 2003 survey conducted on behalf of the National Cattlemen’s Beef Association (1), 188 of 200 processors indicated they used mechanical tenderization to improve
TABLE 4. Direct plating and enrichment results for subprimals inoculated on the lean or fat side with one level of E. coli and then tenderized using a single or a double pass

<table>
<thead>
<tr>
<th>Segment no.</th>
<th>Direct plating*</th>
<th>Enrichment*</th>
<th>Total*</th>
<th>Direct plating</th>
<th>Enrichment</th>
<th>Total</th>
<th>Direct plating</th>
<th>Enrichment</th>
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<td>Total</td>
</tr>
<tr>
<td>1</td>
<td>40/40</td>
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<td>40/40</td>
<td>39/40</td>
<td>1/1</td>
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* Number of segments from which E. coli was recovered by direct plating from among 40 total segments from 40 core samples.
* Number of segments from which E. coli was recovered by enrichment from segments negative by direct plating.
* Number of segments from which E. coli was recovered by both direct plating and enrichment/total sampled (40 total).

product quality. Blade tenderizers work by inserting small needles or double-edged blades into the meat. Regardless of how the subprimals are tenderized, the resulting mechanically tenderized beef steaks are not typically labeled as such, nor are they presently required to be so labeled. Despite studies establishing the low prevalence (4 to 0.2%) of E. coli O157:H7 on the surface of intact meat portions (2), there has been considerable debate about whether there is any added risk of E. coli O157:H7 on the surface of intact meat portions (2). In 1999, the USDA/FSIS also indicated that if nonintact beef products such as needle-tenderized beef became contaminated with E. coli O157:H7, such products would be considered adulterated (16, 17). The debate has intensified recently due to several recalls and illnesses linked to contamination of nonintact meats with E. coli O157:H7. As a result, the USDA/FSIS has informed establishments producing mechanically tenderized beef that were surface inoculated with about 4.0 log CFU/g were statistically (P > 0.05) similar. These data suggest that translocation per inoculum was applied to the fat or lean side of the subprimal were not appreciably influenced by the number of the times the subprimal was passed through the tenderizer. In addition, E. coli O157:H7 cells were recovered from all six core segments obtained from LS, LD, FS, and FD subprimals that were inoculated at ca. 4.0 log CFU/g or cm². Statistically, there was a significantly higher level of the pathogen translocated into segment 1 than into the other five segments for LS, LD, and FD subprimals, but not for FS subprimals. Thus, mechanical tenderization transferred the majority of the E. coli O157:H7 organisms into the topmost 1 cm of the subprimal.

In phase I of this study, in general, for subprimals inoculated with various levels of E. coli O157:H7 and tenderized once on the lean side (LS), the levels of the pathogen recovered from segment 1 (32 to 41%) and the total level recovered from all six segments of a core sample (37 to 55%) were essentially the same, regardless of inoculation level. These results indicate that the level of inoculum that is applied to the surface does not appreciably affect the levels of the pathogen that are transferred into the interior of the subprimal. Likewise, in phase II of this study, levels of the pathogen transferred into segment 1 and the total levels of the pathogen transferred into all six segments of LS, LD, FS, and FD core samples obtained from subprimals that were surface inoculated with about 4.0 log CFU/g were statistically (P > 0.05) similar. These data suggest that translocation per inoculum was applied to the fat or lean side of the subprimal were not appreciably influenced by the number of the times the subprimal was passed through the tenderizer. In addition, E. coli O157:H7 cells were recovered from all six core segments obtained from LS, LD, FS, and FD subprimals that were inoculated at ca. 4.0 log CFU/g or cm². Statistically, there was a significantly higher level of the pathogen translocated into segment 1 than into the other five segments for LS, LD, and FD subprimals, but not for FS subprimals. Thus, mechanical tenderization transferred the majority of the E. coli O157:H7 organisms into the topmost 1 cm of the subprimal.

Table 4 provides the direct plating and enrichment results for subprimals inoculated on the lean or fat side with one level of E. coli and then tenderized using a single or a double pass. The table shows the number of segments from which E. coli was recovered by direct plating from among 40 total segments from 40 core samples. The table also shows the number of segments from which E. coli was recovered by enrichment from segments negative by direct plating. The table further shows the number of segments from which E. coli was recovered by both direct plating and enrichment/total sampled (40 total).
effectively eliminate low levels of the pathogen. Also, if levels of the pathogen were kept relatively low on the surface of meat by following good manufacturing practices and/or by applying interventions to source materials, then presumably there would be fewer cells transferred into the interior upon subsequent blade tenderization. The information herein will be useful in the development of risk assessment models, as well as in the development of safe cooking guidelines, for both blade-tenderized and chemically injected nonintact meats. Studies are ongoing to validate cooking times and temperatures to eliminate cells of *E. coli* O157:H7 transferred to the topmost 1 cm and/or geometric center of blade-tenderized steaks. Likewise, studies are under way to quantify translocation of Shiga toxin–producing *E. coli* into meat that has been enhanced or injected, including any antimicrobial contribution of the brine during injection, shelf life, and/or cooking. Assuming that the prevalence and levels of *E. coli* O157:H7 on the surface of nonintact subprimals remain low and that best practices are followed for operating and monitoring tenderization equipment, then our data and the reports cited herein support the conclusion of others (1, 2, 8, 9, 13, 14) that nonintact, blade-tenderized beef steaks do not present a greater risk to consumers than otherwise similar meat that is intact, provided that the meat is properly cooked.

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**REFERENCES**


