Longitudinal Dissemination of *Salmonella enterica* Clonal Groups through the Slaughter Process of *Salmonella*-Positive Pig Batches

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**ABSTRACT**

This study was conducted to assess the dissemination of *Salmonella* clonal groups in slaughterhouses that received batches of *Salmonella*-positive pigs and used different routine processing procedures. Eight serial sampling sessions were conducted in three slaughterhouses (A, B, and C). Blood was collected randomly (n = 25) from each batch of pigs and processed for serology. Carcasses (n = 12) were identified and sampled after dehairing, after singeing, after evisceration, and before chilling. A section of cecum also was collected. *Salmonella* isolates were submitted to pulsed-field gel electrophoresis. The overall seroprevalence of *Salmonella* was 80.6% (316 of 392 samples), and cecal contents were positive for *Salmonella* in 23.8% (26 of 109) of the pigs sampled. Carcasses after dehairing had a significantly higher prevalence of *Salmonella* (P = 0.004) and the highest *Salmonella* levels (median = 0.25 log CFU/300 cm²). The singeing step significantly affected the *Salmonella* status of the carcasses (P < 0.001); however, the efficacy of singeing differed among slaughterhouses. In the prechilling step, 14.7% (16 of 109) of the carcasses were positive for *Salmonella*. *Salmonella* pulsotypes found on the prechill carcasses were also found in the lairage, in the cecal contents, and on carcasses after dehairing, suggesting that the main source of contamination was the slaughter process before singeing. Slaughterhouse C was the most likely (odds ration [OR] = 6.51) to have pigs carrying *Salmonella* in the gut, and slaughterhouse B was the most likely (OR = 14.66) to have contaminated carcasses at the prechilling step. These findings indicate that the procedures adopted in slaughterhouse B contributed to the spread of *Salmonella* strains. In contrast, in slaughterhouse C the *Salmonella* strains carried by the pigs or found in the lairage were not recovered from prechilled carcasses, validating the effectiveness of the slaughterhouse interventions. These results indicate that an effective slaughter process can help decrease the number of *Salmonella*-positive carcasses in slaughterhouses that receive *Salmonella*-positive pig batches.

The introduction of *Salmonella* into slaughterhouses occurs through *Salmonella*-positive pigs that were infected on the farm, during transport, or at the lairage (3, 8, 9, 20). The major contamination of pig carcasses is pig related (6), and up to 70% of *Salmonella*-positive carcasses are contaminated by carrier animals (4, 8). Although on-farm interventions reduce the prevalence of *Salmonella* in the final products (2–4), the cost-benefit ratio can be high (11); therefore, measures for improving slaughterhouse hygiene and decreasing cross-contamination are needed.

Up to 15% of all carcass contamination occurs during polishing, up to 90% occurs during evisceration, and up to 35% occurs during dressing and splitting (4). Many contamination sources in the slaughter process have been identified, e.g., the intestinal contents and tonsils of infected pigs, the slaughterhouse environment, aerosols produced during carcass splitting, scalding tank water, and dirty pigs entering the slaughter line (5, 6, 8, 9). Differences in processing routines adopted by slaughterhouses at various certain stages can influence the spread of *Salmonella* clonal groups to the carcass during the slaughter process (6, 15). Molecular methods such as macrorestriction profiling by pulsed-field gel electrophoresis (PFGE) have been successfully used to highlight the clonal relationship among strains and determine the routes of contamination in slaughterhouses (7, 9, 27).

Salmonellosis is the most common foodborne disease reported in Brazil (16) and is caused by the consumption of contaminated eggs, chicken, and pork. Batches of pigs with a high *Salmonella* seroprevalence are delivered to some slaughterhouses (21). *Salmonella* has been isolated from fresh pork sausages at the retail level (17) and chilled carcasses (13), highlighting the fact that control measures at slaughter need to be improved. However, to the best of our knowledge no study of *Salmonella* dissemination routes along the pig slaughter line has been conducted in Brazil. This study was conducted to evaluate the dissemination of *Salmonella* clonal groups in slaughterhouses that received *Salmonella*-positive pig batches and that used different routine processing procedures.
MATERIALS AND METHODS

Slaughter process and sample collection. Samples were collected in three commercial slaughterhouses (A, B, and C) that processed ca. 300, 300, and 400 pigs per hour, respectively. A total of eight serial samplings sessions were conducted. Slaughterhouse C was visited twice, and the others were visited three times; the visits were indicated as A1, A2, A3, B1, B2, B3, C1, and C2. The slaughter lines of all slaughterhouses followed a similar procedure, but the main differences were observed for slaughtering, singeing, and cirumcising of the rectum. In all of the slaughterhouses, the pigs were held in lairage for 6 h, stunned with an electric probe, and immediately exsanguinated by severing the carotid arteries and jugular vein. Slaughterhouses A and B had linear scalding tanks, and slaughterhouse C used a scalding shower. The scalded carcasses were dehaired using a rotating drum with a scraper. In slaughterhouses A and C, carcasses were singed in an automated singeing machine. In slaughterhouse B, the carcasses were manually singed with a hand-held gas singer. The singed carcasses were polished by passing them through a series of flails, washed, and then transferred to the evisceration area. Before evisceration (gut removal), in slaughterhouse A a plastic seal was applied around the anus before the rectum was loosened, in slaughterhouse B a string was tied around the distal part of the rectum, and in slaughterhouse C a plastic bag was used to seal off the rectum after it was loosened. The evisceration process was similar in all of the slaughterhouses and included opening of the belly to remove the internal viscera. The carcasses were manually split along the midline with a splitting saw, the blade of which was immersed in hot water before the operation. The head was then removed and the carcass was trimmed, weighed, and washed with cold potable water before chilling.

On each visit, samples were collected when the slaughter process began and continued for 4 h. Samples were taken before slaughtering from the floor of the lairage using the overshoe method, and swabs were taken from the floor of the dressing room at the end of the slaughter period. Scalding water (100-ml) samples were collected in slaughterhouses A and B before slaughter and at 1-h intervals throughout the slaughter process, for a total of five samples per visit. The water temperature was monitored and recorded before each sample was collected.

During the sampling period, pig batches delivered from two or three farms were slaughtered. Blood was randomly collected from 25 pigs per batch during exsanguination and used for serology. After dehairing, the carcasses were identified and swab samples were collected. Sponges were used to swab a 300-cm² area after dehairing, after singeing, after evisceration, and before chilling. Sterile gloves were used for collecting samples and were changed between samples. The swabs were placed in sterile plastic bags containing 50 ml of buffered peptone water (BPW; Merck, Darmstadt, Germany). Samples were collected from at least 12 carcasses on each visit (n = 109). The first carcass was chosen at random after the slaughtering process began, and the subsequent carcasses were chosen at 20-min intervals. A piece of the cecum from each carcass was taken at the time of evisceration. All samples were placed separately in sterile plastic bags and transported to the laboratory in a cooled container for analysis.

Salmonella isolation. Environmental swabs, overshoe, fecal samples (25 g) obtained from each sampled cecum, and scalding water samples (25 ml) were individually transferred to 250 ml of BPW and incubated overnight at 37°C. Plastic bags containing carcass swabs were homogenized, and a 25-ml aliquot was transferred to 250 ml of BPW and incubated overnight at 37°C. Aliquots of this BPW culture were then transferred to both Rappaport-Vassiliadis broth (RV; Merck) and tetraphionate broth (Merck). After incubation at 42°C for 24 h, a 10-μl loop of each culture was streaked onto xylose lysine deoxycholate agar (XLD; Oxoid, Basingstoke, UK) and brilliant green phenol red lactose sucrose agar (Merck) and incubated for 24 h at 37°C. Presumptive Salmonella colonies were confirmed by biochemical and serological tests. The confirmed isolates were serotyped according to the Kauffman-White method at the Brazilian Salmonella Reference Center (Fundação Instituto Oswaldo Cruz, Rio de Janeiro, Brazil).

Salmonella quantification. Swab samples from carcass surfaces were used to quantify Salmonella based on the most-probable-number (MPN) method (25). Plastic bags containing carcass swabs were homogenized, and aliquots (corresponding to 0.1, 1, and 10 ml of the sample) were inoculated in triplicate into 10 ml of BPW and incubated at 37°C for 18 h. The aliquots were transferred to RV (42°C, 48 h, 1:100 dilution) and then plated onto XLD (37°C, 24 h). The number of plates with confirmed Salmonella colonies from each sample aliquot was used to estimate the MPN based on a previously published equation (25).

Salmonella antibody detection. Serum samples were subjected to an indirect lipopolysaccharide–enzyme-linked immunosorbent assay to detect immunoglobulin G anti-Salmonella antibodies. The assay is based on identification of somatic antigens 1, 4, 5, and 12 of Salmonella, representing the serovars most frequently isolated in Brazil, with an optical density cutoff point of 0.169 (14). The herds were classified according to the frequency of positive samples.

Macrorestriction analysis and evaluation of fragment patterns. Salmonella isolates obtained from chilled carcasses and those belonging to the same serovar and originating from the same slaughterhouse were subjected to PFGE analysis (19) following PulseNet procedures (Centers for Disease Control and Prevention, Atlanta, GA; http://www.cdc.gov/pulsenet/protocols.htm). At least two isolates of each sample type were subjected to digestion by XbaI and BlnI (Thermo Scientific Fermentas, Leicestershire, UK). Electrophoresis was performed on a 1% agarose gel using 0.5× Tris-borate-EDTA buffer on a CHEF DR-II system (Bio-Rad Laboratories, Hercules, CA) at 6 V/cm for 20 h at 14°C, with an initial switching time of 2 min and 16 s and a final switching time of 63.8 s. Salmonella Braenderup (ATCC BAA-664) was included as a reference strain. After PFGE, the gel was stained with ethidium bromide (2 μg/ml; Sigma, St. Louis, MO) and photographed under UV transillumination, and the image was digitally processed with a Kodak 2200 system (Kodak, Rochester, NY). Isolates with at least one band difference were considered to be distinct pulsotypes. Pulsotypes were compared using the GelCompar II software package (Applied Maths, Kortrijk, Belgium). Dendograms were constructed using the unweighted pair group method with arithmetic averages (UPGMA) method and the Dice coefficient.

Statistical analysis. All statistical analyses were performed using commercial software (SAS version 9.1, SAS Institute, Cary, NC; PASW Statistics 18, SPSS, Chicago, IL). A significance level of 5% was used. The relationship between seroprevalence and the number of Salmonella intestinal carriers and Salmonella-positive carcasses before chilling in each batch was assessed by nonparametric correlation analysis (Spearman’s correlation). To verify the associations between carcass contamination, Salmonella-positive fecal samples, and the abattoirs (A, B, and C), logistic regression was used. Univariate models with dichotomous
TABLE 1. Frequency of seropositive pigs and Salmonella isolation from cecal contents and prechill carcasses during sampling sessions conducted at three slaughterhouses (A, B, and C) in southern Brazil*

<table>
<thead>
<tr>
<th>Sample session</th>
<th>Serology</th>
<th>Cecal contents</th>
<th>Prechill carcasses</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. positive/no. tested</td>
<td>% positive</td>
<td>No. positive/no. tested</td>
</tr>
<tr>
<td>A1</td>
<td>9/25</td>
<td>36.0</td>
<td>0/12</td>
</tr>
<tr>
<td>A2</td>
<td>17/48</td>
<td>35.4</td>
<td>1/12</td>
</tr>
<tr>
<td>A3</td>
<td>48/50</td>
<td>96.0</td>
<td>3/13</td>
</tr>
<tr>
<td>Total A</td>
<td>74/123</td>
<td>60.2</td>
<td>4/37</td>
</tr>
<tr>
<td>B1</td>
<td>20/42</td>
<td>47.6</td>
<td>2/13</td>
</tr>
<tr>
<td>B2</td>
<td>48/50</td>
<td>96.0</td>
<td>5/13</td>
</tr>
<tr>
<td>B3</td>
<td>53/55</td>
<td>96.4</td>
<td>0/12</td>
</tr>
<tr>
<td>Total B</td>
<td>121/147</td>
<td>82.3</td>
<td>7/38</td>
</tr>
<tr>
<td>C1</td>
<td>63/63</td>
<td>100</td>
<td>13/17</td>
</tr>
<tr>
<td>C2</td>
<td>58/59</td>
<td>98.3</td>
<td>2/17</td>
</tr>
<tr>
<td>Total C</td>
<td>121/122</td>
<td>99.2</td>
<td>15/34</td>
</tr>
<tr>
<td>Total</td>
<td>316/392</td>
<td>80.6</td>
<td>26/109</td>
</tr>
</tbody>
</table>

* Values are number of Salmonella-positive samples/number of samples tested and percentage of positive samples.

The results of Friedman’s test indicated that the levels of Salmonella (log CFU) were not the same among the sampling points on the slaughter line (P < 0.001). Overall,
Salmonella levels reached 0.04 log CFU/300 cm² in slaughterhouse A, whereas in slaughterhouses B and C the levels were 1.24 and 0.32 log CFU/300 cm², respectively. After the singeing step, the 90th percentile Salmonella levels reached 0.04 log CFU/300 cm² in slaughterhouses A and C and 1.10 log CFU/300 cm² in slaughterhouse B. A slight increase in Salmonella levels in the sample steps after evisceration was found in slaughterhouse C.

The analysis of the effect of slaughterhouse on carcass contamination at the different slaughter steps evaluated in this study revealed a higher chance of finding Salmonella isolates (P < 0.05) after dehairing (OR = 4.18), singeing (OR = 11.17), and prechilling (OR = 4.66) in slaughterhouse B than in slaughterhouse A. Slaughterhouse C odds were not significantly different from those of slaughterhouse A. There were higher odds of finding Salmonella isolates in at least one slaughter step (OR = 7.92, P < 0.01) and a higher frequency (OR = 9.24, P < 0.01) of finding Salmonella-positive carcasses at more than one sampling point in slaughterhouse B than in slaughterhouse A.

All samples of the scalding water taken throughout the slaughter period were negative for Salmonella, and the temperature of the scalding water was higher than 61°C at all monitoring points. Among the environmental samples, those taken from the lairage were more frequently Salmonella positive (11 of 14 samples) than were those taken from the dressing room (4 of 16 samples). Salmonella serovars Typhimurium, Agona, Panama, and Derby were isolated from the lairage (Table 2). Salmonella Typhimurium (slaughterhouses A and B) and Salmonella Derby (slaughterhouse B) were recovered from swabs of the dressing room floor.

From the 16 Salmonella-positive carcasses sampled at prechilling, only 2 had been Salmonella negative at all of the previous sampling steps in the slaughter line (Table 2). Of the 10 carcasses that were Salmonella positive at prechilling, 10 were also positive at the dehairing step, 7 were also positive at evisceration, and 4 were also positive at singeing. The number of Salmonella-positive prechill carcasses (16) was equal to or lower than the number of pigs carrying Salmonella in cecal contents (26) in six of the eight sampling sessions. Only four carcasses that were Salmonella positive at the prechilling stage belonged to pigs carrying Salmonella in the cecal contents. The 116 Salmonella strains isolated from carcasses and cecal contents were categorized into eight serovars. Salmonella Derby (43 isolates, 37.1% of the total), Salmonella Typhimurium (41 isolates, 35.3% of the total), and Salmonella Panama (20 isolates, 17.2% of the total) were the most prevalent serovars and the most frequently recovered from the prechill carcass samples. More than one Salmonella serovar was identified in 16 Salmonella-positive carcass samples.

The Salmonella Panama, Salmonella Derby, and Salmonella Typhimurium isolates from five sampling sessions (A3, B2, B3, C1, and C2) for which Salmonella-positive carcasses were identified at the prechilling step were divided into three, six, and eight pulsotypes, respectively, after DNA digestion with XbaI and BlnI (Fig. 3). The similarities between pulsotypes ranged from 35.7% (Salmonella Derby) to 97.6% (Salmonella Panama).

The single Salmonella-positive prechill carcass found in sampling session A3 produced Salmonella Typhimurium strains of two pulsotypes with 88.3% similarity; one of them (T2) was also found in the lairage and another (T1) was not distinguishable from a strain isolated from the same carcass at the dehairing step. In slaughterhouse B, all Salmonella Derby isolates from the carcasses, cecal contents, and environment at sampling session B2 belonged to a single pulsotype, and the Salmonella Panama isolates from two prechilling carcasses at sampling session B3 belonged to the same pulsotype as that of isolates from the lairage and carcasses in the dehairing step. In sampling session C1, one Salmonella Derby isolate from one prechill carcass (5Ch) was similar to isolates from cecal contents (7CC) and two carcasses after dehairing (8Dh and 16Dh), and the isolate from the second prechill carcass (14Ch) was similar to that from cecal contents (4CC and 5CC). In sampling session

**FIGURE 1.** Number of Salmonella-positive carcass samples at the dehairing, singeing, evisceration, and prechilling steps versus the total number of carcasses sampled at Brazilian slaughterhouses A (n = 37), B (n = 38), and C (n = 34).

**FIGURE 2.** Distribution of Salmonella (90th percentile: log CFU/300 cm²) on carcasses at the dehairing, singeing, evisceration, and prechilling steps in three slaughterhouses (A, B, and C) in southern Brazil.
TABLE 2. Salmonella serovars isolated from samples taken from the lairage, pig cecal contents, and pig carcasses at four steps on the slaughter line at three slaughterhouses (A, B, and C) in southern Brazil.

<table>
<thead>
<tr>
<th>Sample session</th>
<th>Lairage</th>
<th>Cecal contents&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Carcasses&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Dehairing</td>
</tr>
<tr>
<td>A1</td>
<td>Typhimurium</td>
<td>Derby (1)</td>
<td>Derby (2)</td>
</tr>
<tr>
<td>A2</td>
<td>Typhimurium</td>
<td>Agona (10)</td>
<td>Typhimurium (2, 12, 13)</td>
</tr>
<tr>
<td>A3</td>
<td>Panama</td>
<td>Panama (11)</td>
<td>Panama (11, 11, 12)</td>
</tr>
<tr>
<td>B1</td>
<td>Typhimurium</td>
<td>Derby (5, 7, 11)</td>
<td>Derby (16)</td>
</tr>
<tr>
<td>B2</td>
<td>Derby</td>
<td>Derby (5)</td>
<td>Derby (10, 12)</td>
</tr>
<tr>
<td>B3</td>
<td>Panama</td>
<td>Panama (11)</td>
<td>Panama (11)</td>
</tr>
<tr>
<td>C1</td>
<td>S. enterica</td>
<td>Derby (5, 7, 11)</td>
<td>Derby (10, 12)</td>
</tr>
<tr>
<td>C2</td>
<td>S. enterica</td>
<td>Derby (5)</td>
<td>Derby (16)</td>
</tr>
</tbody>
</table>

<sup>a</sup> Carcass identification numbers are given in parentheses.
FIGURE 3. Distribution of macrorestriction (PFGE) profiles of the Salmonella serovars isolated from prechilled carcasses from five sampling sessions (A3, B2, B3, C1, and C2) conducted at three slaughterhouses (A, B, and C) in southern Brazil. The dendrograms present the similarity determined by Dice coefficient and UPGMA clustering of the combined XbaI and BlnI profiles. *, Carcass number and origin of isolate. CC, cecal content; Dh, after dehairing; Sg, after singeing; Ev, after evisceration; Ch, before chilling.

Sampling (Origin of isolation*)

B3(11Ev)
B3(Lairage; 11Dh; 11Sg; 11Ch; 12Dh; 12Ch)
B3(12Sg)

A3(12Dh)
A3(5CC)
A3(13Dh; 13Ch)
A3(13Dh)
A3(Lairage; 13Ch)
A3(25Sg; 12CC)
A3(2Dh)
A3(13Dh; Dressing room)

B2(Lairage; 3CC; 4Dh; 4Sg; 4Ev; 4Ch; 5Dh; 5Ev; 5Ch; 6Sg; 6Ch; 7Dh; 7Ev; 7Ch; 8Dh; 8Ev; 8Ch; 9Dh; 9Ch; 10Ev; 11CC; 11Ch; 12Ch; 13Dh; 13Sg; Dressing room)
C1(1CC)
C1(9CC)
C2(12Dh)
C1(4CC; 5CC; 14Ch); C2 (1Ch; 5CC; 9Ev; 10Dh; 12 Dh)
C1(5Ch; 7CC; 8Dh; 16Dh)
C2, the isolates from two prechill carcasses had the same profile as did isolates from cecal contents (5CC) and carcasses after dehairing (10Dh) and evisceration (9Ev).

**DISCUSSION**

A high level of on-farm *Salmonella* transmission was confirmed in pig batches delivered to the three slaughterhouses, based on seroprevalences ranging from 35.4 to 100%. Although high *Salmonella* seroprevalence in herds is thought to increase the number of *Salmonella*-positive fecal samples (15, 22, 23), no correlation was found between the frequency of seropositive pigs and *Salmonella* isolation from cecal samples. Pigs infected on the farm can recover from infection during preharvest, or they can become carriers that excrete *Salmonella* during the holding period (26). Seropositive pig batches comprise both types of pigs, which may contribute to the low level of agreement between the *Salmonella* isolation results from cecal contents and the positive results from the serology tests conducted at slaughter.

Nevertheless, within-herd seroprevalence has been adopted in control programs as an indirect measure of the risk of *Salmonella* introduction by carriers to slaughterhouses (1) and has been correlated with the presence of *Salmonella* on carcass surfaces (15, 22). In our study, only the slaughter batches with a low *Salmonella* seroprevalence (below 50%) had no *Salmonella*-positive prechill carcasses, and seroprevalence of the pig batches was significantly correlated with the number of *Salmonella*-positive prechill carcasses. These results highlight the contamination hazard that the delivery of high-seroprevalence pig batches represents to the slaughter process and slaughterhouse environment, even when no correlation between seroprevalence and *Salmonella* isolation from cecal content is found. The pigs in *Salmonella*-positive slaughter batches can carry *Salmonella* in tissues such as tonsils and lymph nodes, which in turn can be the source of carcass contamination at slaughter (15, 27). Although the presence of carriers is considered the first critical control point (CCP) at slaughter (4), the frequency of *Salmonella*-positive chilled carcasses can be reduced by precautions taken during processing (15, 26). Findings from previous studies suggest that the slaughtering process has a definite impact on the number of contaminated carcasses (1, 23, 26). As in our study, large variations between slaughterhouses in the number of *Salmonella*-contaminated carcasses have been reported, mostly associated with differences in hygienic parameters and control processes (9, 18).

When comparing the different sampling points on the slaughter line, a significantly higher number of positive carcasses was observed after the dehairing process. The high level of contamination before singeing of the carcasses was previously reported to be associated with dirty pigs entering the abattoir (15), contamination from the scalding water (12), or the dehairing process itself (9). In our study, none of the water samples taken from the scalding tank during the slaughtering period were positive for *Salmonella*, and the water was maintained at a temperature above 60 C, which destroys enteric bacteria (5). Therefore, we excluded cross-contamination of carcasses during scalding; however, we cannot assess the contribution of scalding to the reduction in the number of contaminated carcasses because we did not take any carcass samples immediately after this step.

In pigs that carry *Salmonella* in the gut, feces that come into contact with the skin before singeing and during evisceration can spread *Salmonella* over the carcasses (5, 8, 9). However, in our study only four *Salmonella*-positive prechill carcasses originated from pigs that carried *Salmonella* in the gut. These results indicate that most prechill carcass contamination did not occur through contamination of the animal itself, as observed in other studies (4, 27), but mainly through cross-contamination before the singeing step. Singeing was a common, critical step in all of the slaughterhouses studied, although higher mean *Salmonella* counts were found in slaughterhouse B. In this slaughterhouse, the high level of *Salmonella* contamination in the lairage and the inadequate washing of the animals before slaughter may have resulted in pigs with high bacterial counts on their skin entering the slaughter line, as reported by others (5, 15). *Salmonella* pulsotypes found on prechill carcasses were more than 90% similar to those detected in the lairage, cecal contents of other pigs slaughtered on the same day, and carcasses after dehairing, suggesting that the primary source of contamination was the slaughter steps before singeing. Previous researchers have tracked the origin of *Salmonella* strains found on prechill and postchill carcasses to the lairage (9, 10, 13, 23), and the dehairing machine was identified as a major source of bacterial contamination (18). In our study, *Salmonella* clones found in the environment of the lairage and carried in the gut of the slaughtered pigs may have contaminated the dehairing machine through the fecal material escaping from the gut during the dehairing step. Those *Salmonella* clones could then be distributed among subsequent carcasses coming into the contaminated dehairing machine environment.

Singeing also was the most important step for *Salmonella* reduction, contributing to a significant difference (P < 0.001) in carcass status from *Salmonella* positive to *Salmonella* negative. However, the degree of reduction in the number of contaminated carcasses differed among the slaughterhouses and was probably related to the type of singeing process used. Slaughterhouses A and C used an automated singeing system, which resulted in a greater than 80% reduction in the number of *Salmonella*-positive carcasses, whereas slaughterhouse B used a hand-held singeing method that resulted in only a 47.5% reduction. Automated singeing systems have been effective for reducing total bacterial and coliform counts (18, 24) and the number of *Salmonella* isolates (18), although hand-held singeing is thought to be more comprehensively and consistently applied to all areas of the carcass (6). However, in the present study the hand-held singeing method used in slaughterhouse B was not as effective as the automated system for reducing the number of contaminated carcasses or the mean *Salmonella* counts on carcass surfaces. Of the 17 carcasses that were contaminated with *Salmonella* before singeing, 8 remained positive after singeing with the hand-
held unit, whereas only 1 of 13 contaminated carcasses were positive after singeing with the automated system.

Evisceration is considered a CCP for carcass contamination (6) and has been adopted as such in many Brazilian slaughterhouses. The measurement criteria for this CCP are the incidence of gut rupture and the amount of fecal residue on the carcass surface. None of the carcasses sampled in the present study had any visible ruptures or fecal residues, although in slaughterhouses B and C an increase in the number of Salmonella-positive carcasses was noted at this sampling point. Thus, the measurement criteria used for this CCP were not adequate for demonstrating the failure of the slaughterhouse process for targeting Salmonella contamination of carcasses. Singeing has been proposed as a more suitable CCP for pig slaughter (18) and as a cost-effective measure for reducing Salmonella contamination on carcasses (1). Our results support this proposal; the efficacy of singeing had a significant effect on the number of Salmonella-positive carcasses and the Salmonella levels on these carcasses and contributed to a lower number of contaminated prechill carcasses.

Overall, even though pig batches from slaughterhouse C were more likely to carry Salmonella in the gut, the highest odds of finding a contaminated carcass at all sampling points were in slaughterhouse B. The frequency of Salmonella-positive carcasses at the prechilling step was associated only with Salmonella isolates from cecal contents in slaughterhouse B, where the processing steps were inadequate for reducing Salmonella on carcass surfaces. In this slaughterhouse, Salmonella pulotypes were widespread on carcasses sampled at all slaughter points and in the environment. Thus, the procedures adopted in slaughterhouse B contributed to the spread of Salmonella strains, whereas in slaughterhouse C most of the strains carried by the pigs or originating from the lairage were not recovered from the surface of prechill carcasses. In the two slaughterhouses (A and C) that had an effective singeing step, most of the Salmonella strains that were found on the carcasses before the singeing were eliminated by this step. Evisceration and carcasses handling practices in these plants resulted in low levels of recontamination, possibly because an effective good manufacturing practices plan was in place.

The Salmonella seroprevalence of the pig batches was significantly correlated with the number of Salmonella-positive prechill carcasses, indicating that preharvest infection control is crucial for achieving a low prevalence of Salmonella-positive carcasses in southern Brazil. Although on-farm interventions for decreasing the number of Salmonella-seropositive pigs delivered to slaughter is considered the most important stage in Salmonella control programs (1, 15), the time required for these on-farm interventions to reduce Salmonella contamination might be very long in herds with a high Salmonella seroprevalence. In the meantime, high-risk batches of pigs could continue to be delivered to the slaughterhouses and will represent a contamination hazard for the carcasses.

Our results indicated that batches of slaughter pigs with high levels of Salmonella seroprevalence pass on at the presingeing step the Salmonella clonal groups they carry in the gut or on the skin. These clonal groups represent a contamination burden for the presingeing environment, mainly at the dehairing machine, which may be the main site for the spread of clonal groups among carcasses. However, an effective singeing step, in terms of temperature achieved and heat distribution over the carcass surface, can eliminate the Salmonella clonal groups that may have contaminated the carcasses during dehairing. The results of this study indicate that efficiently conducted slaughter processes can contribute to reducing the incidence of Salmonella-positive carcasses, even for high-risk herds of pigs.

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