Variable Abattoir Conditions Affect *Salmonella enterica* Prevalence and Meat Quality in Swine and Pork

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**ABSTRACT**

Research suggests that abattoir holding pens pose significant *Salmonella enterica* risk to swine immediately pre-harvest. The goal of this study was to evaluate those factors related to holding that increased the prevalence of *S. enterica* in swine at slaughter. To accomplish this goal, we focused on holding time and flooring. Our objectives were to (1) compare *Salmonella enterica* prevalence among pigs held for short (15–45 min) versus long (up to 4 h) periods before slaughter; and (2) determine the impact of flooring (slatted vs. concrete) as it relates to the prevalence of *S. enterica*. The study consisted of seven repetitions at a large volume (11,000 head/day) Midwest abattoir. Each repetition consisted of one truck load of pigs (*n*/H11549 170) sorted into one of three groups: (1) animals held for a short time (15–45 min) on solid floors (short-hold); (2) animals held for 4 ± 0.5 h on slatted floors; and (3) animals held for 4 ± 0.5 h on solid concrete floors. At slaughter, samples were collected from 30 pigs in each group. Cecal contents (20 mL), feces (20 g), and the ileocecal lymph node were cultured for *S. enterica*. Additionally, the effect of holding time on meat quality parameters (loin pH at 35 min and 6 h, color, drip loss) was evaluated for the first four replicates. The proportion of *S. enterica*–positive samples was highest (*p* < 0.05) in the cecum of pigs held on solid concrete floors (72.4%), and slightly less for pigs held on slatted floors (63.3%). Animals held for less than 45 min before slaughter demonstrated the lowest proportion of *S. enterica*–positive samples (52.9%). The pig prevalence, as measured by any one of the three samples being positive, was significantly different (*p* < 0.05) between animals held on solid floors (81%) and those animals held for 45 min or less before slaughter (69%). Meat quality, as measured by multiple parameters, was adversely affected by lack of a rest period. The mean 24-h pH was significantly lower for the short-hold group compared to the other two groups. The mean Minolta L and the drip loss were significantly higher in the short-hold group. From this and other studies, it appears that elimination of the holding process is not feasible *S. enterica* control option, given current U.S. harvesting systems.

**INTRODUCTION**

Preliminary research (Hurd et al., 2002) indicates that abattoir holding pens pose a significant *Salmonella enterica* risk to swine immediately pre-harvest. In the United States, market swine are routinely held for 2–6 h after transport and unloading. Holding swine pre-harvest has become an industry-wide practice and is based upon the belief that lactic acid, accumulated during unloading, requires time to dissipate so as to improve meat quality (Warris, 2003). Additionally, the logistics of unloading, weighing, ante-mortem veterinary inspection, and maintenance of sufficient pig flow require holding times of 2–4 h. These holding areas have been shown to contain high levels of environmental *S. enterica* (Rostagno et al., 2003; Swanenburg et al., 2001). Even drinking water sources are often contaminated (Rostagno et al., 2003). Therefore, it has been suggested that reduction or elimination of the

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holding period may be a useful preharvest intervention. Additionally, anecdotal data and pig housing data suggests that the use of slatted flooring might reduce the risk of pig contamination. The objective of this study was to determine if reduced holding times or pens with slatted flooring would affect meat quality and S. enterica recovery rates.

**MATERIALS AND METHODS**

This study consisted of seven repetitions at a large-volume (11,000 head/16-h day) Midwest abattoir. Overall, 630 pigs were tested during seven replicates. Transport time for all replicates was less than 1 h. Pigs were derived from an integrated multi-site production company. Repetitions 1–4 were conducted during the summer months (June to August), while repetitions 5–7 were conducted during the winter months (December to February). Each repetition consisted of pigs from one truckload (n = 170).

During unloading, all pigs were assigned to one of the three groups:

- **Group 1: Short-hold** (15–45 min), where pigs were exposed to a contaminated solid floor not normally used as a holding area
- **Group 2: Slatted hold**, where pigs were held for 4 ± 0.5 h on slatted concrete floors
- **Group 3: solid hold**, where pigs were held for 4 ± 0.5 h on a solid floor. After sorting, the pigs in the short-hold group were weighed, inspected, and walked to stunning as rapidly as the plant operations allowed (15–45 min).

- **Groups 2 and 3**: The slatted and solid groups were moved to their respective pens where they were allowed to rest for 4 ± 0.5 h. Following the rest period, groups 2 and 3 were walked through alleyways to the stunning area, in accordance with the standard plant procedure.

For all three groups, floors were sampled (n = 15) for S. enterica using a four-ply 10 × 10–cm gauze sponge (Nu Gauze, Johnson & Johnson, Arlington, TX) prior to the entry of the study swine. The sorting area and the alleyways were cold water washed before unloading of study pigs. However, the slatted and solid pens were not washed before entry of study pigs, in accordance with normal plant procedures.

Following stunning and evisceration, samples were collected from 30 randomly selected viscera sets in each group. From the viscera sets, cecal contents (20 mL), feces (20 g), and 5 g of mesenteric lymph nodes in the region of the ileocecal junction (ICLN) were collected.

The isolation protocols were as follows: Lymph nodes (average weight 4.6 g, range 1.3–13.8 g) were individually macerated in a sterile bag with a rubber mallet. All media were purchased from Difco Inc (Sparks, MD). Phosphate-buffered saline (25 mL) was added, and each sample was homogenized using a stomacher (Seward, London, UK) for 1 min at 260 RPM. Ten milliliters of supernatant was then added directly to (90 mL) buffered peptone water (BPW) and (90 mL) tetrathionate broth (TB).

Cecal (10 mL) and fecal (10 g) samples were added directly to both BPW and TB (90 mL). BPW and TB were incubated for 24 h at 37°C. Each was then subcultured 1:100 in Rappaport-Vassiliadis broth + novobiocin (RVN; 20 mg/L), incubated for 24 h at 42°C and then transferred 1:100 to Rappaport-Vassiliadis (RV) broth and incubated 24 h at 42°C. RV samples from the BPW pre-enrichment were screened using the Assurance gold EIA Salmonella ELISA kit (BioControl, Bellevue, WA). ELISA-positive BPW samples and all samples from the TB enrichment were then streaked from RV onto XLT4 and modified brilliant green agar (MBG), and incubated for 24 h at 37°C. A single suspect Salmonella colony was then transferred to Tripticase Soy Agar slants (24 h at 37°C) and sent for serotyping to the National Veterinary Services Laboratories (NVSL, Ames, IA).

Standard evaluation protocols were used, by plant personnel, to determine meat quality. These protocols included loin pH, Minolta L color scoring, and JCS. Plant personnel were used as they have daily experience with these techniques. They were blinded as from which
group the samples were derived. The 48-h pH is a critical parameter affecting meat color and texture. Excessively low pH may reflect remaining lactic acid from transport and unloading (Warris, 2003). Loin pH was measured at 35 min and 6 h post-mortem. Final pH (24 h) was determined on the interface of the intact loin after removing a second chop.

Minolta L color scores indicate the redness of the meat and is an important criteria used by American meat buyers (red preferred). The Japanese color score (JCS) is an important color parameter determining whether product quality is sufficient for export. At 24 h post-mortem, color (Minolta L and JCS) and drip loss were assessed on the boneless loin. A 1-inch chop was removed from the sirloin end of the loin and allowed to bloom (set in air for >1 min to allow hemoglobin interaction with oxygen). Objective (Minolta L on cut surface) and subjective (Japanese color score; on cut surface and rib surface) color measurements were obtained after the bloom period. A 1-inch chop was removed from the sirloin end of the loin, and a core sample was obtained from this chop for determination of drip loss (comparison of weight before and after 24 h of drainage).

All data were entered into a spreadsheet, and statistical analysis was performed using JMP version 4 (SAS Institute, Cary, NC). The *S. enterica* isolation rates, for the groups and control groups, were compared for each sample type by a Chi-square analysis. To clearly identify the holding pen’s contribution to slaughter contamination, prior on-farm infections must be identified and excluded from analysis. Therefore, we explored various exclusion criteria in the analysis. Isolates from the ICLN of short-hold pigs could be assumed as originating on the farm. Therefore, serovars found in these tissues were defined as “on-farm” serovars, and the effect of groups were analyzed with and without on farm serovars included. Additionally, only samples positive with serovars recovered from the pen floors before pig placement were evaluated for their effect on group differences.

### RESULTS

The floors in all three areas were highly contaminated, with 95% of the solid, 68% of slatted, and 84% of the short-hold (sorting) area samples testing positive. Pigs unloaded first experienced up to 45 min exposure to these floors in sorting area and alleyways. Table 1 shows the proportion of *S. enterica*–positive samples was higher in pigs held up to 4 h. It was high-

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Lairage condition</th>
<th>All samples</th>
<th>Excluding short hold lymph node serovar</th>
<th>Including only the serovars found in the pen</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cecal contents</td>
<td>Solid hold¹</td>
<td>72.4a (209)</td>
<td>50.9 a</td>
<td>67.32b</td>
</tr>
<tr>
<td></td>
<td>Slatted hold²</td>
<td>63.3a (209)</td>
<td>42.0 b</td>
<td>53.9a</td>
</tr>
<tr>
<td></td>
<td>Short hold³</td>
<td>52.9b (206)</td>
<td>25.2a,b</td>
<td>50.3b</td>
</tr>
<tr>
<td>Fecal contents</td>
<td>Solid hold</td>
<td>23.8a (200)</td>
<td>7.4</td>
<td>22.7a</td>
</tr>
<tr>
<td></td>
<td>Slatted hold</td>
<td>24.7b (197)</td>
<td>9.9</td>
<td>17.1</td>
</tr>
<tr>
<td></td>
<td>Short hold</td>
<td>31.9a,b (188)</td>
<td>16.6</td>
<td>32.4a,b</td>
</tr>
<tr>
<td>Lymph node (ICLN)⁴</td>
<td>Solid hold</td>
<td>34.8 (210)</td>
<td>4.9</td>
<td>30.1a</td>
</tr>
<tr>
<td></td>
<td>Slatted hold</td>
<td>30.9 (210)</td>
<td>5.2</td>
<td>20.8a</td>
</tr>
<tr>
<td></td>
<td>Short hold</td>
<td>35.7 (210)</td>
<td>0.7</td>
<td>27.4</td>
</tr>
</tbody>
</table>

Results within the same sample type and analysis criteria with identical superscripts (a, or b) are significantly different from each other.

¹Pigs held for 4 ± 0.5 h on solid concrete floors.
²Pigs held for 4 ± 0.5 h on slatted concrete floors.
³Pigs waited 15–45 min during sorting and inspection.
⁴Mesenteric lymph nodes in the region of the ileocecal junction (ICLN).
⁵Any sample positive with the same serovar found in the short hold ICLN was excluded from analysis.
⁶Any sample positive with the same serovars found in the pen, before pig placement, were included in the analysis.
est ($p < 0.05$) in the cecum of pigs held on solid concrete floors (72.4%), followed by slatted floors (63.3%), then short-hold (52.9%). The pig prevalence, as measured by any one of the three samples being positive, was significantly different ($p < 0.05$) between solid (81%) and short-hold (69%). When we combined results for the solid and slatted groups to compare long-term to short-term holding, there was a significant difference for the cecal samples (68.2% vs. 53.9%). Analysis, which included only samples positive with serovars recovered from the pen floors before pig placement, resulted in a modest difference for the cecal prevalence among solid, slatted, and short-hold groups (67.2%, 53.9%, 50.3%).

The proportion of positive ICLN was 35.7% in short-hold pigs. Exclusion of serovars found in the ICLN (on-farm serovars) from the analysis increased the differences ($p < 0.01$) between groups for the cecal samples.

Meat quality, as measured by multiple parameters, was adversely affected by lack of a rest period (Table 2). The mean 24-h pH was 5.609 in the short-hold group, significantly lower than the other two groups. Additionally, the mean Minolta L ($49.59$) and the drip loss ($3.619$) was significantly higher in the short-hold group. The Japanese Color Score (JCS) of a cut loin was also significantly lower.

Serovar Typhimurium var. Copenhagen was the most common type recovered: 49.7% of 989 isolates typed. Derby was second, with 35.2% of isolates typed. Few isolates of Anatum (43), Heidelberg (85), Reading (1), Infantis, (3), Newport (1), and Schwarzengrund (14) were also recovered. The mean number of different serovars isolated per replicate was slightly higher in solid (3.15) and slatted (3.0) than short-hold (2.4) pigs.

**DISCUSSION**

In this study, it was not possible to totally eliminate all exposure to contaminated floors due to delays involved in unloading, sorting, weighing, inspection, and walking to stunning. Alleyways in the sorting area still tested salmonella positive after attempts at cleaning and disinfection. Based on other work, it is reasonable to assume that 20–45 min of exposure would increase *S. enterica* recovery rates from the cecal samples (Hurd et al., 2001a). In one study, it was shown that pigs could be internally contaminated in 15 min (Hurd et al., 2001b). As a result, differences in recovery rates between groups was not as large as expected had it been possible to slaughter pigs immediately.

Recovery rates from ICLN were relatively high, compared to expectations of 5–15% from other studies (Hurd et al., 2002). It is possible that there was a relatively high background (on-farm) *S. enterica* prevalence in study pigs. This opinion is supported by the observation that exclusion from the analysis of the serovars found in ICLN increased the observed differences between groups (Table 1).

Important measures of meat quality (loin pH, color, drip loss) were somewhat reduced by eliminating the rest period after transport and unloading. These quality measures are very

<table>
<thead>
<tr>
<th>Quality scores</th>
<th>Short hold</th>
<th>Solid</th>
<th>Slatted</th>
<th>Ideal</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH$^1$</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45 min</td>
<td>6.403</td>
<td>6.369</td>
<td>6.409</td>
<td>6.4</td>
</tr>
<tr>
<td>6 h</td>
<td>6.002$^a$</td>
<td>6.053$^a$</td>
<td>6.129$^a$</td>
<td>6.2</td>
</tr>
<tr>
<td>24 h</td>
<td>5.609$^{a,b}$</td>
<td>5.701$^a$</td>
<td>5.724$^b$</td>
<td>5.8</td>
</tr>
<tr>
<td>Color</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Minolta L$^2$</td>
<td>49.59$^{a,b}$</td>
<td>48.22$^a$</td>
<td>47.12$^b$</td>
<td>47</td>
</tr>
<tr>
<td>JCS–cut$^3$</td>
<td>2.443$^{a,b}$</td>
<td>2.792$^a$</td>
<td>2.875$^b$</td>
<td>3</td>
</tr>
<tr>
<td>JCS–loin$^4$</td>
<td>2.632</td>
<td>2.701</td>
<td>2.681</td>
<td>&gt;3</td>
</tr>
<tr>
<td>Moisture</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Drip loss$^5$</td>
<td>3.619$^{a,b}$</td>
<td>2.269$^a$</td>
<td>2.172$^b$</td>
<td>2</td>
</tr>
</tbody>
</table>

Results within the same sample type and analysis criteria with identical superscripts ($a$, or $b$) are significantly different from each other.

$^1$pH measurement of the loin.

$^2$Instrumental measurement of color (Minolta L*: 0 = darker and 100 = lighter) on the sirloin end cut surface of the loin.

$^3$Subjective score of color on the basis of the Japanese color grid (1 = light/white and 6 = dark/red) on the sirloin end cut surface of the loin.

$^4$Subjective score of color on the basis of the Japanese color grid (1 = light/white and 6 = dark/red) on the rib surface of the loin.

$^5$A measure of the amount of moisture lost from a loin subsample in a 24-h period.

$^6$The ideal represents the meat quality goals of the abattoir where the samples were collected.

The size and current design of most U.S. holding facilities requires that most pigs spend at least 30 min in the contaminated lairage environment for sorting, inspection, weighing, and walking to the stunning chute. Given these constraints and the impact on meat quality, elimination of holding does not appear to be a feasible \textit{S. enterica} control option.

Further research work is suggested from this study. The modestly lower prevalence levels in the ceca of pigs resting on slatted floors (63.3\%) compared to solid floors (72.4\%) suggests some hygiene benefit from this flooring design, which needs further research. The need for a post-transport rest period raises the question of whether pigs could be rested on the trucks. However, this control method would require that pigs spend less than 15 min from unloading to stunning. Additionally, it must be determined whether meat quality is impacted by, for example, transport stress or the physical exertion of unloading. The feasibility of disinfecting pens should be further explored (Schmidt et al., 2004).

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REFERENCES


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