RESEARCH REPORT
PORK SAFETY

Title: National survey of *Salmonella* prevalence in lymph nodes of sows and market hogs – **NPB#16-059**

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Industry Summary:

The primary objective of this study was to benchmark the national prevalence rates of *Salmonella in* the lymph nodes (LNs) of sows and market hogs. LNs were collected across the United States from twenty-one commercial facilities, categorized by geographical regions as north or south. Twenty-five paired superficial inguinal lymph nodes were collected at each facility and shipped to South Dakota State University. *Salmonella* prevalence rates in the northern region were 37.0 and 6.4% for sows and market hogs, respectively. In the southern region, 4.8% of sow samples and 13.0% of market hog samples were *Salmonella* positive. Data on types of chilling methods used at each facility also were collected. In the northern region, prevalence rates of *Salmonella* across chilling types were distributed as follows: 20.0, 2.7, and 1.3% positive samples for conventional, other, and blast chill methods, respectively. Additionally, in the south, there were 20.0% positive samples for conventional, 12.0% for other chill methods, 0.0% for blast.

Keywords: lymph nodes, *Salmonella,* prevalence, sows, market hogs

Scientific Abstract:

Livestock species are known to harbor *Salmonella* in their gastrointestinal (GI) tract and lymphatic tissues. Contamination of carcass surfaces with pathogens harbored in GI tract contents can be mitigated, at least in part, by carcass surface interventions. Lymphatic tissues are generally encased in fat protecting them from carcass surface decontamination treatments, thus serving as a possible root-cause of foodborne illness outbreaks attributed to *Salmonella* in meat products.

By researching the prevalence of *Salmonella* from porcine LNs across the U.S., pork industry members are able to better understand the impact of *Salmonella* as a potential contaminant in pork products. A total of twenty-one commercial pork harvest and processing facilities participated in the study. Facilities were categorized as either north (n = 12) or south (n = 9) geographical regions. As processing volumes allowed, twenty-five carcasses were selected at each establishment, and left and right superficial inguinal LNs (n =1,014 LNs) were removed. For each carcass, left and right LNs were pooled, yielding one sample per animal or n = 507 total LN samples. LNs were then subjected to *Salmonella* prevalence determination. *Salmonella* prevalence rates differed ($P < 0.05$) between hog types in both regions. Specifically, 6.4% of market hog and 37.0% of sow LN samples were found to be *Salmonella*-positive in the north region. This relationship is reversed in the south region as 13.0% of market hog and 4.8% of sow LN samples returned *Salmonella*-positive results. Furthermore, there was a difference ($P < 0.05$) in prevalence rates between regions (north and south) for sows, but not market hogs ($P > 0.05$).

Type of chilling method (conventional, blast, other) used at each facility was documented. Chilling method only relates to the market hogs, as all sow carcasses were hot-boned. In the northern region, prevalence rates of *Salmonella* across chilling types were distributed as follows: 20.0, 2.7, and 1.3% positive samples for conventional, other, and blast chill methods, respectively. Additionally, in the south, there were 20.0% positive samples for conventional, 0.0% for blast, and 12.0% for other chill methods. In both regions, samples from conventionally chilled carcasses returned more ($P < 0.05$) positives results than any other chill method.

Introduction:

According to the Centers for Disease Control and Prevention, *Salmonella* in pork is the third leading cause of foodborne illness-related hospitalizations (Centers for Disease Control and Prevention, 2013). Foodborne illness-related medical costs create an enormous financial burden, even before calculating lost revenue from associated product recalls. For example, data from the United States Department of Agriculture - Economic Research Service, estimates that the annual cost of foodborne illness caused by *Salmonella* in 2013 was $3.7 billion (United States Department of Agriculture - Economic Research Service, 2014). The beef industry has taken advantage of survey type studies to focus on lymph nodes (LNs) as reservoirs for *Salmonella* in raw products. As such, members of the pork industry wanted to assess current prevalence rates of *Salmonella* in the LNs of sows and market hogs in the United States. Therefore, the present study was designed to benchmark current *Salmonella* prevalence rates in those tissues. Data from this study have the potential to influence decisions related to pre- and post-harvest interventions for reducing *Salmonella* in pork, which in turn should reduce the number of salmonellosis cases attributed to pork products. Moreover, controlling *Salmonella* could reduce the amount of regulation changes in the food processing industry.

Objectives:

1. Determine the prevalence of *Salmonella* in the lymph nodes of sows.
2. Determine the prevalence of *Salmonella* in the lymph nodes of market hogs.
3. Determine genetic diversity and outbreak potential of the isolated *Salmonella* strains.
4. Determine the impact of carcass chill method on the prevalence of *Salmonella*.

Materials & Methods:

Sample Collection

Thirty-three commercial pork harvest facilities were originally identified for potential use in this study, and were categorized by hog type (sow or market hog) and geographical region (north or south; Table 1). A total of twenty-one (n = 12 north and n = 9 south) commercial pork harvest and processing facilities agreed to participate in the study; the remaining twelve facilities either declined or are no longer in operation. In-plant LN

collections in the north and regions were conducted by Penn State University and Texas A&M University, respectively.

In addition to LN sample collection, type of carcass chilling method (conventional, blast, or other) used at each facility was documented. Carcass chilling methods were defined as: (1) Conventional – standard cold storage unit without forced air circulation or water spray; (2) Blast chill – cold storage unit with forced air circulation but without water spray; or (3) Other – conventional or blast chill with water spray or other quick chill system. It should also be noted that carcass chilling methods were only documented for establishments harvesting market hogs, as all sow carcasses were hot-boned.

As processing volumes allowed, twenty-five carcasses were selected from each establishment. All samples were collected between the months of December 2016 and August 2017. Left and right superficial inguinal LNs (n =1,014) were removed, and for each carcass, left and right LNs were pooled, yielding one sample per animal or n = 507 total LN samples. Samples were sealed in sterile sample bags, packed in insulated plastic coolers using refrigerant materials, and transported to the Animal Disease Research and Diagnostic Laboratory (ADRDL) at South Dakota State University (SDSU; Brookings, SD) within 24 h of sample collection.

Table 1. Commercial pork harvest facilities by region[a] and hog type

North		South	
Market Hogs (11)	Sows (4)	Market Hogs (10)	Sows (8)
Logansport, IN	Simpsonville, KY	Guymon, OK	Newbern, TN
Beardstown, IL	Xenia, OH	Greenwood, SC*	Union City, TN
Marshalltown, IA*	Watertown, WI	Spring, TX*	Knoxville, TN*
Smithfield, VA*	Des Moines, IA*	Navasota, TX*	Knoxville, TN
Hatfield, PA		Warsaw, NC	Holton, KS
Worthington, MN*		Clinton, NC*	Attala, AL*
Ottumwa, IA		Tar Heel, NC*	Ponotoc, MS
Sioux Falls, SD*		Los Angeles, CA*	Richardson, TX*
Storm Lake IA		Brookshire, TX	
Delphi, IN		Johnson City, TX	
Louisville, KY			

[a] Penn State University and Texas A&M University personnel collected samples from commercial pork harvest facilities in the north and south regions, respectively.
*Commercial Facilities that either declined to participate or were no longer in operation.

Salmonella prevalence and quantification

Once received by SDSU, LNs were aseptically trimmed of fat cover and pulverized, using methods published by Haneklaus et al. (2012). Entire pulverized LN samples (aliquots of pulverized LNs were not used in this study) were analyzed for presence of *Salmonella* using the methods suggested for "raw meat an raw beef mixed products" outlined in the Microbiological Laboratory Guidebook 4.09 – Isolation and Identification of *Salmonella* from Meat, Poultry, Pasteurized Egg, and Catfish Products and Carcass and Environmental Sponges (USDA-FSIS, 2014).

Statistical Analyses

Data were analyzed using JMP Pro Software v13.1.0 (SAS Institute, Inc., Cary, NC). For *Salmonella* prevalence data, contingency tables were produced for region (north, south) and type (market hog, sow), and within-table differences were determined using Fisher's exact test and an $\alpha = 0.05$. To determine differences across chilling methods (conventional, blast chill, other) within a given region, correction for multiple tests was applied to determine significant differences between pairs using an $\alpha = 0.017$.

Results:

Differences in *Salmonella* prevalence by hog type and region are provided in Table 2. Within each region, *Salmonella* prevalence rates between hog types differed ($P < 0.05$). In the north, *Salmonella*-positive

sow samples (37.0%) occurred more often than market hog samples (6.4%). While in the south, a higher rate of *Salmonella* prevalence was seen in LN samples from market hogs (13%) than those from sow carcasses (4.8%). Overall, the rate of *Salmonella* prevalence was higher ($P < 0.05$) in sow samples from the north as compared to those from the south, while the rate of prevalence in market hog samples did not differ ($P > 0.05$) between region.

Table 2. Prevalence of *Salmonella*-positive lymph node (LNs) samples[a] by hog type and region

	Region	
	North	South
Hog type		
Market Hog	6.4 (13/202) A, X	13.0 (13/100) A, X
Sow	37.0 (37/100) B, X	4.8 (5/105) B, Y

A,B: Values within a column lacking a common letter differ ($P < 0.05$).
X,Y: Values within a row lacking a common letter differ ($P < 0.05$).
[a] At the commercial facility, market hogs or sows were harvested and left and right superficial inguinal LNs (n = 1014 LNs) were removed. Within animal, left and right LNs of each type were pooled (n = 507 total samples).

Table 3 shows the rate of *Salmonella* prevalence was highest ($P < 0.017$) for the conventional chill method when compared to other chill types for samples collected in the north (conventional 20.0%; blast chill 1.3%; other 2.7%). No differences in *Salmonella* prevalence were found between chill methods in the south region (conventional 20.0%; blast chill 0.0%; other 12.0%).

Table 3. Prevalence of *Salmonella*-positive lymph nodes (LNs) samples[a] by chilling method[b] and region for market hogs[c]

	Region	
	North	South
Chill type		
Conventional	20.0 (10/50) A	20.0 (10/50) A
Blast chill	1.3 (1/77) B	0.0 (0/25) A
Other	2.7 (2/75) B	12.0 (3/25) A

A,B: Values within a column lacking a common letter differ (P 0.017).
[a] At each commercial facility, market hogs or sows were harvested and left and right superficial inguinal LNs (n = 1,014 LNs) were removed. Within animal, left and right LNs of each type were pooled (n = 507 total samples).

[b] Carcass chilling methods were defined as: (1) Conventional – standard cold storage unit without forced air circulation or water spray; (2) Blast chill – cold storage unit with forced air circulation and without water spray; or (3) Other – conventional or blast chill with water spray or other quick chill system.
[c] Carcass chilling methods were only documented for establishments harvesting market hogs, as all sow carcasses were hot-boned.

Discussion:

In the present study, 13.41% of LN samples confirmed positive for *Salmonella*- (68 positive/507 total samples). Similarly, Pinto-Vieira et al. (2005) found positive samples in the ileum (13.9%), ileocolic lymph nodes (18.8%), tonsils (9.9%), and for the mandibular lymph nodes (12.9%). Several serotypes were identified, with the most prevalent being Typhimurium, Rissen, Tennessee, Enteritidis, Anatum, Give, and Derby (Vieira-Pinto et al., 2005). While these studies provide evidence that porcine peripheral and mesenteric LNs can harbor *Salmonella*, Wang et al. (2010) found 100% *Salmonella*-negative samples after using PCR and cultural methods to analyze 431 subiliac porcine LNs. In a study conducted by Bahnson et al. (2006b), *Salmonella* was not detected in any of 300 pre-scapular LNs analyzed. In the same study, ileocecal lymph nodes were collected from 10 swine herds, and of the 10 herds, 5 tested positive for *Salmonella* (Bahnson et al., 2006b), providing additional evidence that commercial hog populations do harbor *Salmonella* in their LNs.

Generally, literature on *Salmonella* prevalence in porcine peripheral LNs is limited, especially with regard to regional differences. Data from the present study revealed regional differences in *Salmonella* prevalence. To date, there is no known literature available regarding *Salmonella* prevalence in the southern U.S., although studies conducted in the north have been documented. O'Connor et al. (2006) conducted a study on finishing swine in Iowa. In the first part of the study, 5,054 diaphragm samples were tested from low-volume producers. Resulting in 38.97% (1,863/5,054) samples testing positive for *Salmonella*. In addition to evaluating low-volume producers, diaphragm samples were collected from market hog lots of high-volume producers. For each lot, *Salmonella* prevalence is as follows; 60% (27/45 herds), 27% (12 herds), 11% (5 herds) and 2% (1 herd) (O'Connor et al., 2006). Furthermore, Bahnson et al. (2006a) investigated *Salmonella enterica* prevalence from ileocolic LNs of hogs in Midwest swine herds. Researchers found *Salmonella* in 100 of the 146 herds sampled (68.5%), with individual-pig prevalence of 6.98% positive within-herd (Bahnson et al., 2006a). These data demonstrate the potential for the high prevalence of *Salmonella* in hog herds in the north region.

Larsen et al. (2003) conducted a study to determine the prevalence of *Salmonella* in cull sows using many different tissue types, including ileocecal, ventral thoracic, and subiliac LNs. Of the 181 samples collected, 12 ileocecal, 4 ventral thoracic, and 4 subliac LNs were positive for *Salmonella*, resulting in an overall *Salmonella*-prevalence rate of 8.84% (Larsen et al., 2003).

Data from the current study demonstrate a higher *Salmonella* prevalence rate in samples from conventionally-chilled carcasses as compared to other chilling methods. Bahnson et al. (2006a), found that freezing samples to -70°C, did not result in decreased *Salmonella* prevalence in ilocolic LNs (Bahnson et al., 2006a). Conventional chill could be considered the slowest method of the three chilling types evaluated. Therefore, this might play a role in the increased *Salmonella* prevalence.

Overall, findings from this study provide the U.S. pork industry with a valuable set of benchmark data that establish levels of *Salmonella* prevalence in the peripheral LNs of market hog and sow carcasses. These results have the potential to influence decisions related to pre- and post-harvest interventions for reducing *Salmonella* in pork, which in turn should reduce the number of salmonellosis cases attributed to pork products. Additional research opportunities might also be identified to further explore the impact, if any, that rate of chilling and type of chilling may have on *Salmonella* prevalence in porcine LNs.

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