NATIONAL SURVEY OF *SALMONELLA* PREVALENCE IN LYMPH NODES OF SOWS AND MARKET HOGS

A Thesis

by

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ABSTRACT

Livestock are known to harbor *Salmonella* in their gastrointestinal (GI) tract and lymphatic tissues. Pathogens on carcass surfaces can be mitigated by antimicrobial interventions applied to the surface. Lymph nodes (LNs) are typically below the surface and encased in fat protecting them from typical antimicrobial treatments, thus serving as a possible root-cause of foodborne illnesses attributed to *Salmonella* in meat products. To establish a baseline of *Salmonella* prevalence in porcine LNs across the U.S., twenty-one commercial pork harvest and processing facilities, representing northern (n = 12) or southern (n = 9) geographical regions, participated in this study. As processing volumes allowed, twenty-five carcasses were selected from 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. *Salmonella* prevalence was determined for all samples. *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 northern region. This relationship was reversed in the southern 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 northern and southern regions for sows, but not market hogs ($P > 0.05$). Type of chilling method (conventional, blast, or other) used at each market hog facility (n = 12) was documented. 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, combined, and blast chill methods, respectively. Additionally, in the southern region, 20.0% of samples were

positive for conventional, 0.0% for blast, and 12.0% for other. In both regions, samples from conventionally chilled carcasses returned more ($P < 0.05$) positive results than any other chill method. Results of this study provide a much-needed baseline for *Salmonella* prevalence rates in LNs of sows and market hogs in the U.S.

DEDICATION

I dedicate this work to my family and friends. Without their constant love and support I wouldn't be where I am today.

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This work was supervised by a thesis committee consisting of Professor Jeffrey Savell [co-advisor] and Professor Kerri Gehring [co-advisor], and Professor Davey Griffin, and Research Assistant Professor Ashley Arnold of the Department of Animal Science.

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NOMENCLATURE

LN	Lymph Node
CDC	Centers for Disease Control and Prevention
USDA	United States Department of Agriculture
FSIS	Food Safety and Inspection Service
GI	Gastrointestinal Tract
H	Hours
SDSU	South Dakota State University

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1. INTRODUCTION

According to the Centers for Disease Control and Prevention (2016b), *Salmonella* causes approximately 1.2 million illnesses and 450 deaths every year in the United States. *Salmonella* is the second leading cause of foodborne illness in the United States, and the leading cause of foodborne illness hospitalizations and death in 2015 (Centers for Disease Control and Prevention, 2016a). Furthermore, the most common food/pathogen pairs for foodborne illnesses in 2015 were seeded vegetables/*Salmonella* (1,048 illnesses), pork/*Salmonella* (615), and *Salmonella*/vegetable row crops (263). From the same report, the most documented food/pathogen pairs that caused the most hospitalizations were as follows; seeded vegetables/*Salmonella* (225 hospitalizations), pork/*Salmonella* (70), and chicken/*Staphylococcus aureus* (31) (Centers for Disease Control and Prevention, 2015). In 2015, the United States Department of Agriculture - Food Safety and Inspection Service (2014) issued a public health alert for a Class One Recall of approximately 116,262 pounds of whole pigs used for pig roasts after 134 case patients were identified in Washington. Foodborne illness-related medical costs create an enormous financial burden, even before calculating lost revenue from associated product recalls. For example, the United States Department of Agriculture - Economic Research Service (2014) estimates that the annual cost of foodborne illness caused by *Salmonella* in 2013 was $3.7 billion.

Lymph nodes (LNs), and the lymphatic system are a part of the body's immune system to collect fluid, waste, viruses, and bacteria that are in the body's tissues (The American Cancer Society, 2015). Lymphatic tissue and LNs have been identified as a source of *Salmonella* because LNs can still harbor infections within them. Generally, the majority of studies regarding LNs have been conducted on LNs located in the gastrointestinal tract. Nevertheless, *Salmonella*

has been identified in peripheral LNs that have the potential to be incorporated in ground products. *Salmonella* in peripheral LNs becomes an issue because they are protected from carcass interventions due to the surrounding fat tissues. There are 31 pathogens known to cause foodborne illness (Centers for Disease Control and Prevention, 2016a). Therefore, the lymphatic system will sequester such bacteria, and begin the process of ridding the body of the harmful microorganisms. This creates the potential for *Salmonella* to be harbored in the LNs.

When considering the medical costs potentially associated with *Salmonella* in LNs, the need for research to reduce associated illnesses is emphasized. The beef industry has taken advantage of survey-type studies to focus on LNs as reservoirs for *Salmonella* in raw products. Due to the number of illnesses and previous data reporting that bovine lymph nodes harbor *Salmonella*, 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.

II. REVIEW OF LITERATURE

Salmonella Prevalence

Kampelmacher et al. (1963) investigated the epidemiology of *Salmonella* infections in pork in the Netherlands and found a correlation between *Salmonella* infections and pork products. They found 181/600 (30.1%) *Salmonella*-positive samples from a variety of porcine sources, including: crura of diaphragm (5.5%), spleen (3.1%), liver (3.9%), gallbladder (9.6%), mesenteric LNs (15.0%), portal LNs (8.0%), and feces (11%). From 181 positive animals, the most prevalent *Salmonella* serovars were: *S.* Typhimurium (67%), *S.* Heidelberg (30%), and *S.* Bredeney (17%), (Kampelmacher et al., 1963). This study provided a general baseline for *Salmonella* prevalence and associated serovars within pork products in the Netherlands. Data from this study show that there was an issue of internal and external contamination of market hogs (Kampelmacher et al., 1963). In a more recent study, Duffy et al. (2001) determined prevalence of microbial contamination in U.S. pork products. They collected a total of 384 samples of retail pork and found *Salmonella* in 9.6% of samples collected. Their data showed that ground products had a higher incidence of contamination when compared to retail products (Duffy et al., 2001). In a similar study, Swanenburg et al. (2001) collected a large number of carcass samples: *Salmonella* occurred in 25.6% of rectal content samples, 19.6% of tonsils, 9.3% of mesenteric LNs, and 1.4% of carcass swabs. Moreover, *Salmonella* was isolated from one or more samples in 47% of the pigs (Swanenburg et al., 2001). Data from Swanenburg et al. (2001) show that the prevalence of *Salmonella* in slaughtered pigs can be high, and that LNs can be a source of *Salmonella* in pork products. Similarly, Pinto-Vieira et al. (2005) found positive samples in ileocolic (18.8%) and mandibular LNs (12.9%). Pinto-Vieira et al. (2005) also documented a number of serovars from these samples, with the most prevalent being

Typhimurium, Rissen, Tennessee, Enteritidis, Anatum, Give, and Derby. Data from Vieira-Pinto et al. (2005) indicate LNs being more sensitive for detection of *Salmonella* prevalence when compared to other tissue types.

Extensive *Salmonella* prevalence data have been collected from different sample types, including feces, organs, and LNs. Hurd et al. (2001) evaluated the effects of lairage on *Salmonella* from market hogs at slaughter and collected a variety of samples for *Salmonella* prevalence testing. The authors found *Salmonella* in 71.8% for all sample types collected (ceacal and distal colon contents, and ileocecal LNs). Of these samples, 43.6% of LNs tested positive for *Salmonella,* with Agona, Derby, and Typhimurium being the most prevalent serovars. These findings led the authors to believe that peripheral LNs could be a significant source of *Salmonella* contamination (Hurd et al., 2001). In a separate study conducted by Hurd et al. (2002), they looked at *Salmonella enterica* prevalence within herds regarding transport distance and holding times. The authors collected superficial inguinal and ileocecal LNs at harvest, pooled them, and determined an overall rate of 9.1% *S. enterica* prevalence at harvest, further demonstrating that LNs located outside the gastrointestinal tract can harbor *Salmonella.* Similarly, a study was conducted by Kim et al. (1999) on 30 swine herds to assess the relationship between *Salmonella* prevalence on-farm and at harvest. Of 966 LNs collected at harvest, 13.7% tested positive for *Salmonella*. All of these data provide evidence that *Salmonella* in LNs at harvest pose a potential food safety risk.

While the aforementioned studies provide evidence that porcine peripheral and mesenteric LNs can harbor *Salmonella*, Wang et al. (2010) conducted a study to assess 30 feces and 30 subiliac LNs (n=60) from 24 farms as predictors of *Salmonella* prevalence in live hogs. The researchers found 3.4% of farm feces and 0.06% of subiliac LNs were *Salmonella*-positive.

Nevertheless, 71.4% of herds tested positive for *Salmonella* in one or both of the samples collected. The most prevalent serovar isolated from *Salmonella*-positive LNs was Braenderup, while the dominant on-farm serovars were Derby, Anatum and Typhimurium (Wang et al., 2010). Conversely, in a two-part study, Bahnson et al. (2006b) researched the prevalence of *Salmonella* in prescapular LNs from market swine. In part one of the study, no *Salmonella* was detected in 300 prescapular LNs collected. In the second part of the study, ileocecal and prescapular LNs were collected from 10 swine herds containing 75 pigs each. Like the first part of the study, *Salmonella* was not detected in prescapular LNs. Nevertheless, 5 of 10 herds tested positive for *Salmonella* in the ileocecal LNs. Derby, Typhimurium, Java, Hartford, Mbandaka, and Senftenberg serovars were isolated from ileocecal LNs (Bahnson et al., 2006b). While these results are difficult to compare to our present work, they provide additional evidence that commercial hog populations do harbor *Salmonella* in their LNs.

Regional Prevalence in the United States

According to United States Department of Agriculture - Animal and Plant Health Inspection Service (1995), "salmonellosis in swine should be viewed as two separate problems. First, as a disease in swine causing septicemia and diarrhea, and second as a potential source of contamination in pork carcasses and retail cuts." In 1995, the researchers (United States Department of Agriculture - Animal and Plant Health Inspection Service, 1995) gathered information on *Salmonella* prevalence in on-farm fecal samples from pork farms around the United States, ultimately finding 38.2% *Salmonella*-positive samples. Furthermore, they showed 65.5, 36.1, and 29.9% for the southeastern, northcentral, and midwest regions, respectively (United States Department of Agriculture - Animal and Plant Health Inspection Service, 1995). O'Connor et al. (2006) conducted a two-part study on *Salmonella* antibodies by collecting

diaphragm samples from low- and high-volume Iowa pork producers. Of samples tested, 18.9% of hog lots from low-volume and 19.7% of hog lots from high-volume producers tested positive for *Salmonella* antibodies (O'Connor et al., 2006). The producers were classified by having negligible, low, moderate, or widespread evidence of historical exposure to *Salmonella.* While low-volume producers displayed negligible or low evidence of exposure, implications from this study show that high-volume producers have a higher occurrence of *Salmonella* positive market hogs (O'Connor et al., 2006). While this study evaluated *Salmonella* antibodies, it exhibits how many hogs are exposed to *Salmonella* while on farm. Additionally, Bahnson et al. (2006a) investigated *Salmonella enterica* prevalence from ileocolic LNs of hogs in midwest swine herds. *Salmonella* was found in 100 of the 146 (68.5%) herds sampled (Bahnson et al., 2006a). These data demonstrate the potential for high prevalence rates of *Salmonella* in hog herds in the northern region.

Regional Prevalence in Europe

Even though few data are available on regional-differences in the United States, there have been several studies conducted in European countries to evaluate *Salmonella* prevalence. As a follow-up study to Kampelmacher et al. (1963), Edel and Kampelmacher (1976) conducted a study at 7 harvest facilities located throughout the Netherlands. To make this study comparable to the original, they collected portal and mesenteric LNs and feces from 700 hogs harvested at 7 facilities. The *Salmonella* positive results from this study were as follows, facility one (75%), two (45%), three (40%), four (24%), five (13%), six (17%), and seven (24%). The authors concluded that despite then-current food safety practices, *Salmonella* was still occurring (Edel and Kampelmacher, 1976). In Canada, Lammerding et al. (1988) isolated *Salmonella* from 17.5% of pork neck muscle, portal LNs, and mesenteric LNs. Moreover, *Salmonella*

brandenburg was the most predominant serovar, followed by Derby, Agona, Infantis, Copenhagen, Anatum, Kentucky, London, Muenster, Typhimurium, Bredeney, Choleraesuis, Heidelberg, Ohio, and Schwarzengrund (Lammerding et al., 1988). More recently, there was a study conducted on herd prevalence of *Salmonella enterica* infections in Danish slaughter pigs by Baggesen et al. (1996). Overall, 6.2% of pigs were positive for *Salmonella enterica*, with the predominant serovar being *S.* Typhimurium (64.4%) (Baggesen et al., 1996). In total, there were 30 different serovars identified. These data show low *Salmonella* prevalence rates as compared to other countries, including the United States. Furthermore, Kashbohrer et al. (2000) conducted an epidemiological study on market hogs in Germany. The authors collected a total of 36,000 samples that consisted of fecal, LN and carcass swabs from seven abattoirs located throughout Germany. *Salmonella* was isolated from 3.3% of LNs, and the estimated overall prevalence of *Salmonella* was 6.2% in slaughter pigs. Data for the LNs displayed significant differences between facilities in 14 and 17 of the 21 harvest facilities for LNs and fecal samples, respectively. More recently, Jung et al. (2001) conducted a study in South Korea for *Salmonella* prevalence in 784 ileocecal LNs from harvest pigs. They found 140 (17.9%) *Salmonella*-positive LNs samples, containing the following serovars: Typhimurium (41/784), Derby (20/784), Schwarzengrund (23/784), Mbandaka (19/784), Enteritidis (6/784), Agona (6/784), Braenderup (3/784), Newport (4/784), Ruiru (4/784), Rissen (3/784), Litchfield (2/784), Tennessee (1/784), Kinshasa (2/784), Eimbsebuettel (2/784), and Havana (1/784) (Jung et al., 2001). It is important to note that this study supports LNs as a viable source of *Salmonella* contamination, and some serovars found have caused foodborne illnesses. That same year, Camitz et al. (2001) conducted a study on *Salmonella* antibodies in swine for the HerdChek enzyme-linked immunosorbent assay (ELISA) kit. The Dutch Animal Health Service implemented the HerdChek kit to detect

Salmonella antibodies in the pork production system in Germany and compared it to the United States *Salmonella* antibody rate. In the United States 22.7% of samples were positive for *Salmonella*, and 56.7% of the samples collected in Germany were positive for *Salmonella (Camitz et al., 2001)*. While the HerdChek kit has been implemented to locate *Salmonella* antibody occurrence in pork production so interventions can be updated to reduce *Salmonella*, this shows there is a difference among geographical locations. Finally, Lo Fo Wong et al. (2003) collected data on fecal samples for *Salmonella enterica* prevalence within 77 herds from Denmark, the Netherlands, Greece and Germany. Overall, 42% of herds were *Salmonella*-positive. Additionally, Germany had 5% of herds test positive for *Salmonella,* and the only serovar detected was Derby; 23.5% of herds from Greece tested *Salmonella* positive, with the serovars Typhimurium, London and Bredeney; 85.0% of fecal samples from Denmark tested positive for *Salmonella,* and the only serovar detected was Typhimurium; 50% of herds in the Netherlands tested positive for *Salmonella,* with positive samples consisting of serovars Typhimurium, London, Bovismorbificans and I, O21, nm. These data show regional differences within European countries, with Denmark having the highest rate of *Salmonella* (Lo Fo Wong et al., 2003). Therefore, from these data we can conclude there is the possibility for regional differences within the United States.

Hog-Type

Data regarding impact of gender on *Salmonella* prevalence in LNs are limited. Larsen et al. (2003) conducted a study to determine the prevalence of *Salmonella* in cull sows ileocecal, ventral thoracic, and subiliac LNs. Of the 181 samples collected, 12 ileocecal, 4 ventral thoracic, and 4 subiliac LNs were positive for *Salmonella*, resulting in an overall *Salmonella*-prevalence rate of 8.8% (Larsen et al., 2003). While the main focus of this study was on holding and lairage,

the usage of only sows provides helpful gender-specific information. The United States Department of Agriculture - Animal and Plant Health Inspection Service (1995) conducted a national study on *Salmonella* prevalence producing some data on sex-type differences. According to this study, single-sex pens (25%) were twice as likely to contain pigs shedding *Salmonella* than mixed pens (12.7%). These data show some sex-type differences in *Salmonella* prevalence. van der Wolf et al. (2001) conducted a baseline study on the *Salmonella* prevalence in hogs in the Netherlands. ELISA was used to detect *Salmonella* antibody prevalence for finisher hogs, free-range finishers, sows, and gilts. Gilts produced 0% antibody-positive samples, but sows produced 60.4% antibody-positive samples for *Salmonella.* The finishers were 24.5% antibody-positive, and the free-range finishers were 44.6% antibody-positive. There were significant differences between sows and gilts, and between finishers and free-range finishers. From these data, the Netherlands can identify herds with a high *Salmonella* prevalence and assess production practices to reduce the amount of *Salmonella.*

Chilling Methods

Additional research is needed on chilling applications and associated *Salmonella* prevalence in pork. Bahnson et al. (2006a) found that freezing samples did not result in decreased *Salmonella* prevalence in ileocolic LNs. Conventional chilling could be considered the slowest method of the three chilling styles evaluated in our study. Therefore, this may play a role in the increased *Salmonella* prevalence. Nevertheless, Vanantwerpen et al. (2016) studied the effect of chilling method on *Salmonella* recovered from hog carcasses. Two harvest facilities were chosen, facility A had a fast cooling system with airflow, and B had a conventional cooling system with no airflow. This study showed that chilling system did not significantly affect the recovery of *Salmonella* on pork carcasses swabs (Vanantwerpen et al., 2016). The studies

mentioned above show that chilling methods do not impact the prevalence of *Salmonella.* Still, more research is needed to fully understand the relationship, or lack thereof, between chilling method and *Salmonella* prevalence in LNs.

Salmonella Prevalence Monitoring Systems

The Danish *Salmonella* surveillance-and-control program is applied to every step in the Danish pork system to create an efficient way of identifying herds that have high *Salmonella* prevalence. They do so by implementing regulations at feeding, breeding, weaning, finishing and harvest (Nielsen et al., 2001). The classification system was implemented in 1995, and according to Nielsen et al. (2001), the program has reduced *Salmonella* prevalence and the number of salmonellosis cases attributed to Danish pork from 1993 to 2000 (Nielsen et al., 2001). Likewise, Alban et al. (2002) reviewed the classification system that was implemented by the Danish *Salmonella* surveillance-and-control program in 2000. There were five main points that were addressed and corrected for this program to make the classification system more efficient: (1) sampling systems simplified, (2) eliminating small herds from surveillance, (3) adjusting cut-offs for detection level, (4) weight of the 3 previous months to assign a *Salmonella* prevalence level more quickly, and (5) a herd being assigned monthly to one of three levels (Alban et al., 2002).Through these monitoring systems, a country can assess its *Salmonella* prevalence rate and adjust production practices accordingly.

Pathogen reduction is a major objective for the meat industry in the United States. Therefore, in 1996, the United States Department of Agriculture's Food Safety and Inspection Service (USDA-FSIS) implemented the Pathogen Reduction; Hazard Analysis and Critical Control Point System. This established performance standards for *Escherichia coli* and *Salmonella* (United States Department of Agriculture - Food Safety and Inspection Service,

1996). A few years later, in 1998, the USDA-FSIS implemented new performance standards for *Salmonella* (United States Department of Agriculture - Food Safety and Inspection Service, 1998). More recently, The United States Department of Agriculture - Food Safety and Inspection Service (2018) has implemented the proposed New Swine Slaughter Inspection System (NSIS), requiring additional pathogen sampling for all swine establishments. The NSIS is optional for swine harvest facilities, allows for faster line speeds, and more off-line inspection practices. All of this can happen because they are implementing new antemortem inspection practices, and facilities will perform on-line inspections and trimming before the inspector. Through this new system, facilities would be allowed to adjust their inspection and sampling procedures to fit their harvest facility. Therefore, the pork industry would have the ability to implement systems much like the Danish surveillance-and-control system. Moreover, different harvest practices could be implemented to reduce *Salmonella* prevalence, such as removing major lymph nodes so they do not end up in ground product.

III. MATERIALS AND METHODS

Lymph Node Collection

Thirty-three commercial pork harvest facilities were initially identified as potential participants and were categorized by hog type (sow or market hog) and geographical region (northern or southern). A total of twenty-one (n = 8 northern market hog, n = 4 northern sow, n = 4 southern market hog, and n = 5 southern sow) commercial harvest and processing facilities participated in the study; the remaining twelve facilities either declined or were no longer in operation. In-plant LN collections in the northern and southern regions were conducted by Penn State University and Texas A&M University personnel, 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 another quick chill system. Carcass chilling methods were only documented for establishments harvesting market hogs, as all sow carcasses were hot-boned.

Sample Collection and Processing

Twenty-five carcasses were selected from each establishment, except for one sow facility with a low processing volume and one market hog facility where two extra carcasses were sampled. All samples were collected between December 2016 and August 2017. From each carcass, the left and right superficial inguinal LNs (n =1,014) were removed and pooled, yielding one sample per animal or n = 507 total LN samples. Samples were sealed in sterile sample bags

(VWR International; Randor, PA), packed in insulated hard plastic coolers with refrigerant materials, and transported within 24 h of sample collection to the Animal Disease and Diagnostic Laboratory (ADRDL) at South Dakota State University (SDSU; Brookings, SD). Upon arrival, LNs were aseptically removed from surrounding fat tissue using flame-sterilized scalpel and forceps. De-fatted LNs were flame-sterilized to remove any surface contamination, weighed, placed into sterile filter bags (Whirl-Pak, Nasco, Sandy Springs, GA), and pulverized using a rubber mallet. Pulverized LN samples were stored in refrigerated conditions (~ 4 °C) overnight until microbiological analyses were performed

Salmonella Isolation and Confirmation

LN samples were pre-enriched with 90 mL of buffered peptone water (BPW) and incubated for 18 to 14 h at 37 °C. Entire samples (aliquots of pulverized LNs were not used in this study) were analyzed for presence of *Salmonella* spp. using the methods suggested for "raw meat and raw beef mixed products" according to the procedures outlined in the Microbiological Laboratory Guidebook 4.08 – Isolation and Identification of *Salmonella* from Meat, Poultry, Pasteurized Egg, and Catfish Products and Carcass and Environmental Sponges (United States Department of Agriculture - Food Safety and Inspection Service, 2014).

Salmonella enterica in pulverized LN samples was detected using real-time PCR method (BAX System; DuPont Qualicon, Wilmington, DE) by screening the overnight enrichment for the presence of *Salmonella* DNA. The samples that were either BAX PCR positive or indeterminate were cultured by transferring 1 mL of pre-enriched cultures into 10 mL Tetrathionate broth (BD Difco; Sparks, MD) and subsequently incubating at 37 °C for 18 h. After incubation, cultures were streak plated onto selective Xylose-Lysine-Tergitol 4 agar (BD Difco) and incubated at 37 °C for 24 h. Plates that exhibited black colonies after 24 h incubation

were presumptively classified as *Salmonella enterica* and stored at 4 °C. These colonies were further sub-cultured in Luria-Bertani agar plates overnight at 37 °C and verified for *Salmonella* using a Bruker Matrix-Assisted Laser Desorption Ionization – Time of Flight Mass Spectrometer.

Statistical Analysis

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 (northern, southern) and hog 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, combined) within a given region, Bonferroni's correction for multiple tests was applied to determine significant differences between pairs using an $\alpha = 0.017$.

IV. RESULTS AND DISCUSSION

Differences in *Salmonella* prevalence by hog type and region are provided in Table 1. 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%). Conversely, in the south, a higher rate of *Salmonella* prevalence was seen in LN samples from market hogs (13%) than from sow carcasses (4.8%). Overall, the rate of *Salmonella* prevalence was higher ($P < 0.05$) in sow samples from the northern as compared to those from the southern region, while the rate of prevalence in market hog samples did not differ ($P > 0.05$) between region.

Table 1. Prevalence of *Salmonella*-positive lymph node (LNs) samples[1] by hog type and region

	Region	
Hog type	North	South
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 ($P < 0.05$).
[1] A total of twenty-one ($n = 8$ northern market hog, $n = 4$ northern sow, $n = 4$ southern market hog, and $n = 5$ southern sow) commercial harvest and processing facilities participated in the study; the remaining twelve facilities either declined or were no longer in operation. 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).

As previously stated, Larsen et al. (2003) determined 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 subiliac LNs were positive for *Salmonella*, resulting in an overall *Salmonella*-prevalence rate of 8.8% (Larsen et al., 2003). The present study displayed a *Salmonella* prevalence for market hogs and sows to be 20.5% and 8.6%, respectively. These values were higher than the prevalence documented by Larsen et al. (2003).

The United States Department of Agriculture - Animal and Plant Health Inspection Service (1995) stated that hogs from the southeastern United States had a higher prevalence of *Salmonella* than the midwest and northcentral regions. This differs from findings of the present study. The differences seen between this present study and the United States Department of Agriculture - Animal and Plant Health Inspection Service (1995) study could be due to environmental differences or the time of year. 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 hog herds sampled, with an overall prevalence of ~7.0% positive for the 4,380 collected (Bahnson et al., 2006a). Bahnson et al. (2006a) displayed a higher overall prevalence for *Salmonella* than was seen in our study.

Table 2 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%; combined 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%). These data 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 ileocolic LNs (Bahnson et al., 2006a). Conversely, Vanantwerpen et al. (2016) found that chilling system did not significantly influence the recovery of *Salmonella.* While findings from Vanantwerpen et al. (2016) and (Bahnson et al., 2006a) are in agreement with our results from the southern region, our northern region data did show a significant difference for conventional chill.

Table 2. Prevalence of *Salmonella*-positive lymph nodes (LNs) samples[1] by chilling method[2] and region for market hogs[3]

	Region	
Chill type	North	South
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 ($P < 0.017$).

[1] A total of twenty-one (n = 8 northern market hog, n = 4 northern sow, n = 4 southern market hog, and n = 5 southern sow) commercial harvest and processing facilities participated in the study; the remaining twelve facilities either declined or were no longer in operation. 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).

[2] 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.

[3] Carcass chilling methods were only documented for establishments harvesting market hogs, as all sow carcasses were hot-boned.

V. CONCLUSION

This survey was conducted to establish a benchmark for the national prevalence rates of *Salmonella spp.* in the LNs of sows and market hogs, and the findings provide valuable information for its presence in the peripheral LNs of sows and market hogs. These data serve as an introduction of different effects (region, sex, chilling method) of *Salmonella* prevalence, and gives direction to future studies conducted by the pork industry.

There were implications from these findings that must be taken into consideration by pork producers and processors. Further research is needed to identify production practices contributing to *Salmonella* in LNs. Specifically, items for consideration include development and implementation of on-farm production practices, veterinary treatments, and pre-harvest interventions. Additionally, developing and implementing post-harvest processing methods to reduce *Salmonella* in porcine LNs should be explored more thoroughly. Because LNs are a possible source of *Salmonella* contamination, pre-harvest practices and/or procedures for removing LNs during processing may be beneficial in reducing *Salmonella* in pork, thereby reducing foodborne illnesses.

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