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Environmental Prevalence and Persistence of *Salmonella* spp. in Outdoor Swine Wallows

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ABSTRACT

Swine can harbor *Salmonella* in their gastrointestinal tracts. It has been estimated that up to 48% of the U.S. swine herd may carry *Salmonella*. Housing sows in farrowing stalls has become controversial due to animal welfare-based criticisms. An alternative production system is to keep sows outdoors on pasture with access to individual farrowing huts. This study was designed to determine the effects of two production systems on indicator bacteria and *Salmonella* of sows housed indoors in farrowing stalls ($n = 52$) compared to sows housed outdoors ($n = 52$) in English style huts. Each farrowing radial contained one wallow, from which mud ($n = 290$) and water ($n = 290$) samples were collected weekly. All samples were analyzed for generic *E. coli*, coliforms and *Salmonella*. No differences ($p > 0.05$) were detected in *Salmonella*, generic *E. coli* and coliform populations between indoor farrowing stalls and outdoor farrowing huts. However, all 8 outdoor wallows contained *Salmonella* spp. at some point during the study ($n = 49$ *Salmonella* isolates). *Salmonella* genotypes persisted within some wallows for >5 months, and genetically indistinguishable *Salmonella* isolates were found in multiple wallows. *Salmonella* isolated from outdoor sow feces were genetically indistinguishable by PFGE from *Salmonella* isolated from wallows ($n = 33$) throughout the study, indicating that pathogenic bacteria were cycling between swine and their environment. In conclusion, the role of wallows in disseminating *Salmonella* within an outdoor swine herd appears to be significant.

INTRODUCTION

SWINE CAN BE ASYMPTOMATIC reservoirs of food-borne pathogenic bacteria that are transmissible to humans via consumption of contaminated pork products or through the environment (Davies et al., 1999; Rostagno et al., 2003). Food-borne pathogenic bacteria, such as *Salmonella*, are found in the environment of pig farms (Funk et al., 2001; Letellier et al., 1999), and can persist in the environment or within a herd at a subclinical levels for years (Sandvang et al., 2000). It has been estimated that between 25% and 48% of the U.S. swine herd may be colonized with *Salmonella* species on the farm (Davies et al., 1997; Funk et al., 2001).

Swine are traditionally raised in the U.S. under intensive confinement conditions, with

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more than 80% of sows and piglets housed in farrowing crates (NAHMS, 2000). However, the farrowing stall has received criticism due to potential detrimental effects it may inflict on the well-being of the sow (HSUS, 2004). An alternative system is housing the sow and her litter on pasture with access to an individual farrowing hut; and this system has been promoted (HSUS, 2004) as a sustainable system that enhances animal welfare. However, caution should be taken in only considering swine welfare when advocating any production system. Other important considerations include effects upon environmental and food safety.

To date, no comparison between these two intensive swine production systems has been made in regards to the carriage or shedding of food-borne pathogenic bacteria. Therefore this study was designed to compare the prevalence of fecal contamination indicator bacteria (i.e., coliforms and generic *E. coli*) and *Salmonella* in the farrowing environment of sows and piglets housed indoors individually in farrowing stalls versus those housed outdoors in English-style huts, and secondly to determine the persistence of fecal indicator bacteria and *Salmonella spp*. in outdoor farrowing wallows.

MATERIALS AND METHODS

Animals

Breeding age gilts were obtained from a single source farm and were considered to have a high health status. Research was conducted at the Texas Tech University Sustainable Pork Farm situated in an area with a dry steppe climate producing mild winter temperatures near Lubbock, Texas. Average weather data during this study are given in Table 1 (NWS, 2002). A total of 108 sows ($n = 54$ indoor and $n = 54$ outdoor; Newsham genotype, Colorado Springs, CO) and their litters were used in this study.

Diets, housing, and husbandry

The research was conducted over a 10-month period (March 2001 to January 2002) at two farms. Animals were housed and used in accordance with FASS regulations (FASS, 1999) and the project was approved by the Texas Tech University Animal Care and Use Committee.

Sows in both environments remained outdoors or indoors completely during breeding, gestation, and farrowing. Within indoor and outdoor gestation groups sows were of mixed parity and all sows were artificially inseminated. Outdoor sows were kept in the same social group through both the gestation and farrowing phases. In the indoor facilitates there were 16 farrowing stalls per room and for outdoor sows groups contained eight sows per group per farrowing paddock. Five days before their scheduled farrowing date all sows were transferred to their respective farrowing facilities, either farrowing pasture with individual farrowing huts (outdoors) or farrowing stalls (indoors).

Table 1. Weather Measurements for the Sustainable Swine Farm Located in Lubbock County, Texas, from March 2001 to January 2002

Climatic measurement	*Mar*	*Apr*	*May*	*Jun*	*Jul*	*Aug*	*Sep*	*Oct*	*Nov*	*Dec*	*Jan*
Air temperature, °C											
Avg	9.06	17.89	21.17	27.06	29.61	26.50	22.17	16.44	11.33	5.83	5.67
Avg min[a]	2.78	9.61	13.61	19.00	22.06	19.50	14.72	7.67	5.22	−1.28	−2.39
Avg max[b]	15.28	26.22	28.72	35.06	37.17	33.5	29.61	25.17	17.44	12.94	13.72
Monthly min[c]	−3.33	3.89	6.67	13.33	18.33	16.11	7.78	−1.11	−7.78	−6.11	−8.89
Monthly max[d]	27.78	33.33	35.00	40.56	40.56	37.22	35.56	32.22	28.89	20.00	28.11
Total precipitation, cm	6.22	0.97	10.67	1.19	1.52	2.82	2.16	0.05	8.76	0.33	1.57
Average wind velocity, km/h	18.24	23.2	20.00	21.92	17.60	14.56	16.64	19.52	18.40	18.24	19.04

[a]Average minimum daily temperature, °C.
[b]Average maximum daily temperature, °C.
[c]Average minimum relative humidity, %.
[d]Average maximum relative humidity, %.

Eight 0.4-ha radial paddocks containing English-style arc farrowing huts were used in this study to house outdoor farrowing sows and their litters. Short chopped wheat straw was used for bedding. Fenders were attached after the sow had chosen her farrowing hut and prior to piglets being born (Johnson and McGlone, 2003).

Sows were fed once daily at 0800 in a designated grassy strip area along one side of the perimeter fence with a completely balanced sorghum-based diet (CP 19.9%). A continuous dewormer was added to the diet (Banminth 48, Pfizer, Lee Summit, MS). Depending on the stage of lactation, sows in both environments were fed the appropriate amount of feed meal (NRC, 1998). Outdoor piglets were not provided with a creep feed but did have access to ground cover continuously during the summer months.

One wallow per farrowing paddock was built prior to sow arrival. Initial wallow dimensions were 5.4 m × 0.90 m × 0.3 m. A clean supply of drinking water was provided to sows via a perforated PVC tube suspended over each wallow. With water continually circulating through the wallow, water remained algae-free and wallows always contained adequate water rather than a thick mud layer.

Indoor sows were housed in conventional farrowing stalls. Waste fell into a pit and was removed 2–3 times/day by water flushing. Sows were fed once a day at 0800 with a completely balanced sorghum-based diet (CP 19.9%) in a metal trough. A nipple drinker system supplied continuous water to both sows and piglets. Room temperature was kept at 27–29°C. Indoor piglets were provided a heat lamp for the first 7 days and, at 14 days of age, a creep feeder containing a pre-starter/early wean diet (Metabalance 10/15 CP 22%, Consolidated Nutrition, Quincy, IL) was added to the pen (Johnson, 2001).

Comparison of the prevalence of generic E. coli, *coliforms, and* Salmonella *in the farrowing environment for sows and piglets housed indoors versus outdoors*

This study was conducted over a period of ten months ($n = 10$ months). One hundred ($n = 100$) environmental samples from indoor sows and their litters and 100 environmental samples from outdoor sows and their litters were collected (Table 2) during monthly samplings ($n = 10$/environment/month). Samples were examined for determination of *Salmonella* via enrichment and quantitative determination of fecal-contamination indicator bacterial species (generic *E. coli* and total coliform populations) by serial dilution and direct plating. Drinking water (50 mL; $n = 1$/environment/month) from the first water from the nipple system (indoor) and PVC drip water samples (outdoor wallows) and feed samples (50 g; $n = 1$/environment/month) immediately prior to feeding on site were collected in sterile 50-mL

Table 2. Sample Locations and Types for Indoor and Outdoor Intensive Swine Production Facilities

	Monthly samples from indoor facility	*Monthly samples from outdoor facility*	*Total number of samples collected*
Water 50 mL	1	1	20
Feed 50 g	1	1	20
Environmental			
Sow belly swab	2	2	40
Piglet belly swab	2	2	40
Stall floor	2	—	20
Stall wall	2	—	20
Fender roller wall	—	2	20
Used straw from hut floor	—	2	20
Feces pooled from 5 sows	1	1	20
Wallows			
Mud	—	Weekly	290
Water	—	Weekly	290

conical tubes (Becton Dickson Labware, Franklin Lakes, NJ) tubes.

Homologous environmental structures (e.g., fender rollers and farrowing stalls) and bellies of sows and piglets in both facilities were sampled once per month ($n = 160$ total samples; 8 samples/environment/monthly sampling). Samples of the environment of both the facilities and animals were collected using a 10 cm × 10 cm sterile cotton swab, soaked in sterile 0.9% saline solution. Swabs were wiped over a surface using a sterile 20 × 20 cm area template that was sterilized with 70% ethanol between samples; swabs were placed into separate sterile resealable plastic bags immediately after swabbing and stored on ice for 24 h during transport. Locations swabbed included: bellies of sows (both indoor and outdoor; $n = 2$ swabs/environment/month), bellies of piglets (both indoor and outdoor; $n = 2$ swabs/environment/month), sow stall floor (indoor only; $n = 2$ swabs/month) and stall wall swabs (indoor only; $n = 2$ swabs/month), and hut fender roller walls (outdoor only; $n = 2$ swabs/month), and, used straw was collected (outdoor only; $n = 2$ samples/month) from farrowing hut floors and placed into sterile resealable plastic bags.

Fecal samples (>20 g) were collected from five randomly selected sows each month in each facility (indoor and outdoor) placed in separate sterile plastic bags at the time of collection. Monthly fecal samples were pooled (10 g sub sample/sow) and stored on ice for 24 h during transport to FFSRU in College Station, TX for bacterial analyses.

Bacterial persistence in wallows

To determine the persistence of bacteria in the wallow environment, samples of mud ($n = 8$ samples weekly; $n = 290$ samples total) and water ($n = 8$ samples weekly; $n = 290$ samples total) were collected weekly over the course of 37 weeks from the wallow within each radial ($n = 8$) at the outdoor intensive facility. In five ($n = 5$) cases, feces were collected from the wallow mud/water interface in five separate wallows as an additional sample. These fecal samples were only collected if they could be clearly identified as a fresh, defecation from a single animal that had not been mixed with the mud or water by the passage of sows or piglets. These samples were collected to minimize mud/water contamination of the sample.

Sample preparation

Sterile Brilliant Green Bile Broth (BGB; Oxoid Ltd., Basingstoke, UK) was added in a 1:9 ratio to resealable plastic bags containing all swabs and water to dilute material recovered from the environment and animal hides, and bags and swipes were mechanically mixed (via Stomacher) for 1 min to thoroughly mix each swab sample and diluent (hide swab diluent). Fluid from this initial swab dilution was used immediately for further dilutions to estimate *E. coli* and total coliform populations as well as for qualitative enrichments for *Salmonella*.

Feces (10 g), feedstuffs (10 g), drinking water (10 ml), wallow mud and water samples (10 g or 10 mL) were individually weighed and added in a 1:9 ratio to bags containing sterile phosphate buffered saline (PBS; pH 7.0); samples were mechanically mixed for 1 min to mix each sample and diluent thoroughly. Fluid from this initial dilution was used to quantitatively enumerate generic *E. coli* and total coliforms. To enrich *Salmonella* populations, the materials were diluted (1:9) and qualitatively enriched as described below.

Quantitative determination of generic E. coli *and total coliforms*

Fecal, feedstuffs, wallow mud and wallow water samples were serially diluted (in ten fold increments) in PBS for enumeration of total coliforms and generic *E. coli*. Enumerative dilution series (10–fold increments) were plated directly onto MacConkey's Agar (to enumerate total coliforms), and M-Endo LES agar (for enumeration of generic *E. coli*). Colonies were directly counted on plates after 24 h of incubation at 37°C.

Qualitative enrichment of salmonella

For qualitative enrichment of *Salmonella*, feces (3 g), feedstuffs, mud or 3 mL of water or hide swab diluent (in BGB) were added to tubes containing 27 mL of Tetrathionate broth (Difco Laboratories) and incubated at 37°C for

24 h. After this incubation, 200 μL of the Tetrathionate enrichment were added to 5 mL Rappaport-Vassiliadis R10 broth (Difco, 1998) and incubated an additional 24 h at 42°C before being streak-plated onto brilliant green agar (BGA) supplemented with novobiocin (25 μg/mL; BGA_{Nov}). The BGA_{Nov} plates were incubated for 24 h at 37°C; colonies that exhibited typical *Salmonella* morphology (pink/white, opaque, circular, entire, convex colonies of a medium size surrounded by a brilliant red medium [alkaline]) were individually picked for further biochemical characterization. Positive (*S*. Typhimurium) and negative (*E. coli*) control cultures were grown in parallel with samples to ensure quality of agar. Picked putative *Salmonella* colonies were inoculated onto Triple Sugar Iron (TSI) agar slants and Lysine Iron agar (LIA) slants (Difco, Inc.). Each slant was incubated at 37°C for 24 h. *Salmonella*-positive samples were confirmed by slide agglutination using SM-O antiserum poly A-I and V-I, and group C1 factors. *Salmonella* isolates were stored in glycerol and TSB at −80°C until confirmatory serotyping was performed by the National Veterinary Services Laboratory (NVSL) in Ames, IA.

Pulsed-field gel electrophoresis (PFGE)

PFGE was performed using the standard Food Safety Inspection Service (FSIS) protocol for subtyping *Salmonella* with minor modifications (Wonderling et al., 2003). Confirmed *Salmonella* isolates (by NVSL serotyping; $n = 23$) from outdoor wallow mud and water samples were selected as representative of all wallow *Salmonella* isolates and were subjected to PFGE analysis. Briefly, *Salmonella* isolates were grown on TSA plates for 16 h at 37°C and suspended in buffer (100 mM Tris, 100 mM EDTA) to an OD_{610} of 1.25 ± 0.1. An equal volume of 1.4% Low Melt agarose, 1% SDS containing 1 mg/mL proteinase K (Invitrogen, Carlsbad, CA) was added. Aliquots were dispensed into disposable 0.1 mL plug molds (Bio-Rad, Hercules, CA) and allowed to solidify for 20 min. Plugs were transferred to 50 mL conical bottom tubes containing 5 mL lysis buffer (50 mM Tris, 50 mM EDTA, 1% sarcosine, 0.1 mg/ml proteinase K) and incubated at 54°C for 2–4 h in a shaking water bath. Plugs were washed 2× in H_2O, 2× in 0.5M TBE (650 mL each wash) in a PVC plug washer for 30 min each wash step. Plugs were sectioned into thirds and digested with 50 U XbaI (Invitrogen) for 6 h at 37°C. Plugs were incorporated into 1% Pulsed Field Grade Agarose gel (Bio-Rad) and pulsed field electrophoresis was performed using a CHEF Mapper XA system (Bio Rad) in 0.5X TBE at 14°C with pulses ramping from 2.16 to 63.8 sec over 18 h. Fragment size was compared against a standard control strain of *Salmonella* per CDC protocols (CDC, 1998). The gel was stained with ethidium bromide, visualized, and stored as a TIFF file (ChemiDoc, Bio Rad). Analysis was conducted using Bionumerics software (Applied-Maths, Austin, TX). Cluster analysis using the Dice correlation for band matching with a 1% position tolerance and hierarchic UPGMA was used to generate a dendrogram describing the relationship of *Salmonella* isolates.

Statistical analysis

For coliform and generic *E. coli* population data from indoor and outdoor environments, a Student's *t*-test was performed to compare means for each sample type, and significance differences were determined at $p < 0.05$.

RESULTS

Comparison of the prevalence of E. coli, *coliforms, and* Salmonella *in the farrowing environment for sows and piglets housed indoors versus outdoors*

Drinking water and feed samples contained no *Salmonella* at any time during the study. Only one drinking water sample during the entire study contained any generic *E. coli* or coliforms; however feed samples from both facilities consistently contained 10^1 to 10^4 CFU/g feed of coliforms and generic *E. coli* (Fig. 1). Data from both farms are pooled and presented in Figure 1.

Weather conditions during the course of this study were typical for a North Texas year (Table 1). Swabs collected from the walls and floors of stalls and huts as well as the belly of sows and piglets all contained both coliforms

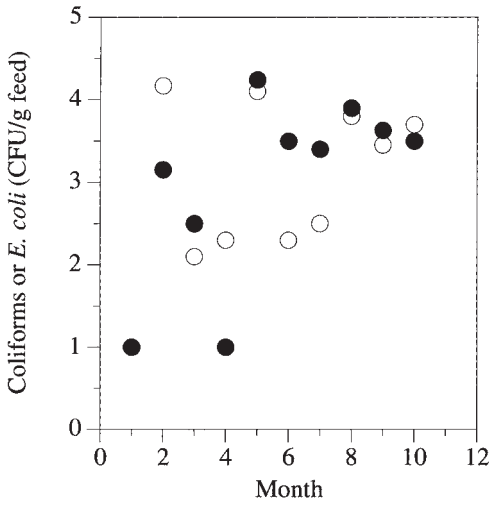

FIG. 1. Coliform and *E. coli* populations in feed (CFU/g) from both indoor and outdoor intensive swine farms. Open symbols represent coliform populations, and shaded bars depict *E. coli* populations.

and generic *E. coli* at concentrations from 10^4 to 10^6 CFU/swab (Fig. 2a,b). There were no differences ($p > 0.10$) detected between swabs taken from the indoor or outdoor facility for either environmental or swine belly swabs. Fecal populations of coliforms and *E. coli* were 10^6 to 10^8 CFU/g feces, but again there was no significant difference ($p > 0.10$) between production facilities (data not shown).

All *Salmonella*-positive samples from the indoor were isolated directly from sow feces (n = 6 pooled fecal samples, representing 30 individual sows, from the 10 monthly samplings). *Salmonella* serotypes isolated from feces from the indoor facility were: Derby, Give, Kentucky, Meleagridis, and Muenster. Samples of straw collected from the floor of a single farrowing hut at the outdoor facility contained *Salmonella* Give only on two ($n = 2$ samples from the 10 monthly samplings) separate sampling dates.

Determination of the persistence of E. coli *and* Salmonella *spp. in a wallow*

Both the water and mud in wallows in all radials contained coliforms and *E. coli* and means from all 8 wallows are presented in Figures 3a and 1b. Bacterial populations in water from the wallows were variable, but were not ($p > 0.05$) significantly different over time (Fig. 3a). Mean bacterial populations in the mud from the wallows remained relatively constant throughout the study, at a level of approximately 10^{4-5} CFU generic *E. coli* and coliforms/g mud (Fig. 3b).

Salmonella spp. (n = 45 isolates, representing 4 serotypes) were isolated from the wallows in every radial over the course of this study (Table 3). *Salmonella* isolates were found in wallow mud (n = 27 *Salmonella* positive samples out of 290 mud samples) and wallow water (n = 18 *Salmonella* positive samples out of 290 water samples). The most common serotype isolated was *S*. Give (33 isolates), followed by *S*. Mbandaka (10), *S*. Derby (1), and *S*. Typhimurium (1). *S*. Give appeared in at least one sample from all radials except one (radial 7), and was detected at least intermittently for >3 months in 3 radials (1, 2, and 4). *S*. Mbandaka was isolated at least once from four of the eight radials during this survey. PFGE results from 23 se-

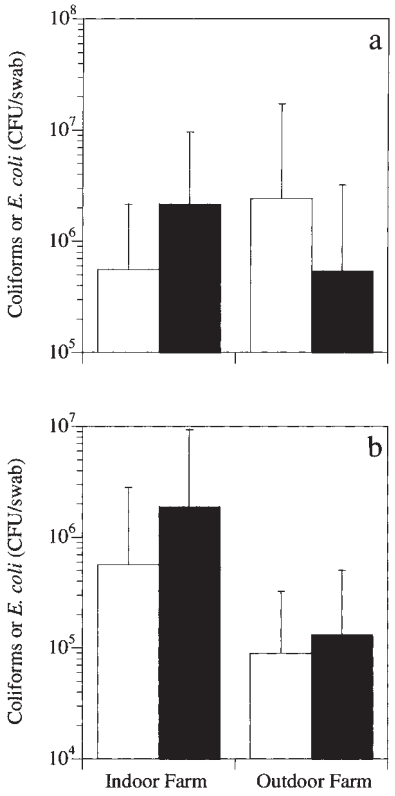

FIG. 2. Coliform and *E. coli* populations (CFU/swab) from indoor and outdoor intensive swine farms. Environmental swabs from within farrowing huts and farrowing crates (**a**), and swabs from the bellies of sows and piglets (**b**). Open bars represent coliform populations, and shaded bars depict *E. coli* populations.

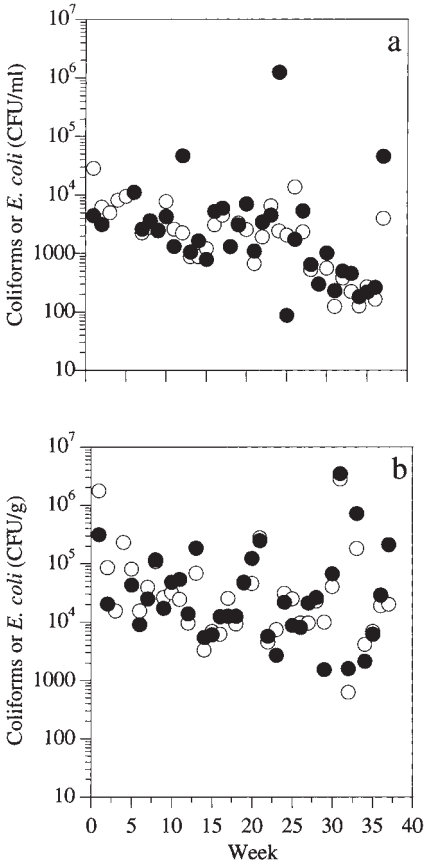

FIG. 3. Populations (CFU/mL or g) of coliforms and generic *E. coli* from outdoor wallow water (**a**) and mud samples (**b**). Open symbols represent means (n = 8 wallows) coliform populations, and filled symbols indicate generic *E. coli* populations.

lected *Salmonella* spp. isolates indicated that the *S*. Give isolates from all 7 radials in which it appeared, and across a 5-month period within a single radial were isolates of a clonal origin (Fig. 4). Additionally, the *S*. Give isolated from swine manure deposited on the edge of the wallow in radial 2 (Table 3) was genetically indistinguishable from all other *S*. Give isolates from this study (Fig. 4). The *S*. Mbandaka isolates from all radials in which it appeared, as well as isolates from the same radial across a 3-month period were also clonal with respect to each other (Fig. 2).

DISCUSSION

The U.S. pork industry, like other animal production commodity groups, is being challenged on a variety of issues, including the environment, antibiotic use, and animal welfare and rights (Berends et al., 2001; Edwards and Zanella, 1996; EU, 1997). One animal care challenge has focused on the type of housing used for the sow and her litter at the time of farrowing and lactation. The majority of sows in the EU and the United States (83.4 ± 4.0 %) are currently housed in farrowing stalls (NAHMS, 2000). The farrowing stall gained popularity among producers for many reasons—they are a cost effective use of space, easy to clean and garner better worker safety. However, the farrowing stall has also raised some animal welfare concerns, especially in regards to a limitation on allowing the sow movement (HSUS, 2004). An alternative option to use of the farrowing stall is intensive outdoor pasture farrowing. Sows farrow and nurse their litters in small huts, floored with straw, but sows can freely leave the hut and piglets. Wallows are provided in each paddock to help alleviate heat stress during the hotter summer months.

Pathogen dissemination into the environment and food supply is of great interest when discussing swine housing and potential effects on the quality and safety of pork products. *Salmonella* spp. infections annually cause more than 1.3 million human illnesses, at a cost to the U.S. economy of more than $2 billion (ERS/USDA, 2001). *Salmonella* species are commonly isolated from swine around the world (Davies et al., 1997; Sandvang et al., 2000; Weiss et al., 2002), and the on-farm incidence of *Salmonella* in swine in U.S. operations is estimated at 38% (NAHMS, 1997).

A previous investigation of the connection between swine production systems and foodborne pathogens found that multiple-site all-in/all-out management strategies had no benefit in reducing the prevalence of *Salmonella* in swine compared with more traditional farrow-to-finish operations (Davies et al., 1997). It was found that the prevalence of *Salmonella* was highest in swine finished in dirt lots; however, the production systems examined (Davies et al., 1997) were different than the system utilized in the present study. The prevalence of *Salmonella* from environmental samples (from facilities, bellies and feces) within the farrowing stall housed sows in the current study was

TABLE 3. *SALMONELLA* SEROTYPES ISOLATAED FROM MUD (N = 27 ISOLATES) AND WATER (N = 18 ISOLATES) SAMPLES FROM INDIVIDUAL FARROWING WALLOWS

	Radial													
	1		*2*		*3*		*4*		*5*		*6*	*7*	*8*	
Sampling date	*Mud*	*Water*	*Mud*	*Water*	*Mud*	*Water*	*Mud*	*Water*	*Mud*	*Water*	*Mud*	*Mud*	*Mud*	*Water*
5.14				Derby										
5.21	Give		Give				Give							Give
5.30	Give	Give						Give						
6.06				Give							Give			
6.13														
6.20			Mbandaka	Mbandaka		Mbandaka								
7.02				Mbandaka										
7.12			Give											
7.18									Give	Give				
7.31							Give							
8.22		Give		Give			Give							
8.29	Give		Give (fecal)	Mbandaka		Mbandaka								
9.05		Give	Give	Give			Mbandaka		Mbandaka					
9.17			Give	Give	Mbandaka									
9.24			Mbandaka	Give	Give								Give	
10.01			Give											
10.24			Give				Give							
10.30			Give				Give	Give	Give					
11.06							Give							
1.10												Typhimurium		
Total isolates	3	3	10	9	2	2	7	2	3	1	1	1	1	1

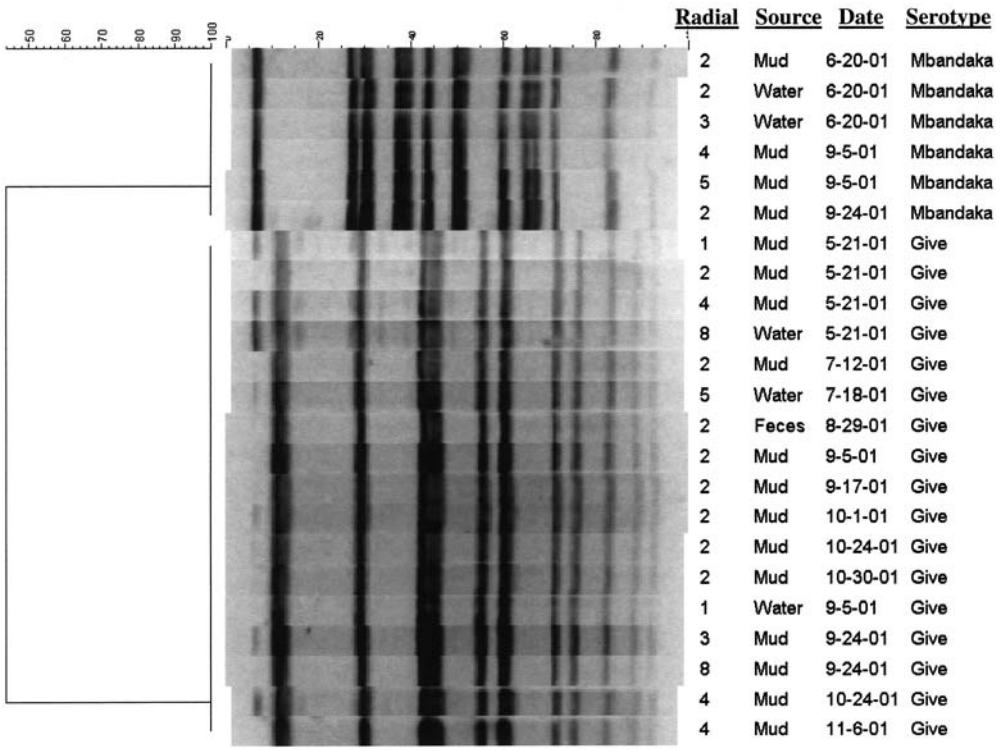

FIG. 4. Pulsed-field gel electrophoresis gel (PFGE) of selected *Salmonella* isolates from outdoor wallows described in Table 2 over the period of the study. Dendrogram ruler indicates percentage of genetic relatedness of isolates based on PFGE profile.

less than 7%, and the prevalence rate of homologous samples from the outdoor farm was approximately 2%. Given such very low *Salmonella* prevalence in the indoor and outdoor facilities, no conclusions about food safety impact of the two intensive swine production systems can be drawn from our data.

Pigs may become colonized with *Salmonella* by ingesting contaminated feces; however, esophogatomized swine can quickly (< 3 h) become colonized with *Salmonella* following intranasal inoculation, suggesting that the respiratory system and/or tonsils could facilitate colonization (Fedorka-Cray et al., 1995). Placing swine in *Salmonella*-contaminated pens at slaughter plants for a lairage period can quickly (2–4 h) result in the colonization of pigs with multiple serovars (Hurd et al., 2001; Rostagno et al., 2003). Other research has indicated that one of the greatest risk factors for transmission of *Salmonella* within pens or herds is snout-to-snout contact (Lo Fo Wong et al., 2004). Therefore, the fecal-nasal or fecal-oral route of contamination plays an important role in *Salmonella* transmission amongst swine (Proux et al., 2001; Winfield and Groisman, 2003).

In the present study, pigs were observed to root in, and drink water from the wallows. This potential route of oral or intranasal contamination is obviously of significance, especially given the fact that fresh feces collected from near a wallow contained *Salmonella* Give that was genetically indistinguishable from all of the *S*. Give isolates from the wallows throughout the study. Although *S*. Give has rarely been implicated in human food-borne illness outbreaks, it has been isolated from food animals (CDC, 2003; Roy et al., 2001), and has been associated with several food-borne illness outbreaks around the world (EFSA, 2004). It cannot be determined if the *S*. Give contaminated wallow was the initial source of contamination, or if another source was responsible. The role of piglets or other animal vectors carrying *Salmonella* between radials or re-inoculating each wallow could not be determined in the present study, and a sampling of insects and transient animals within the radials yielded no *Salmo-*

nella isolates (data not shown). Based on the literature, it seems likely that at least some of these vectors play a role in *Salmonella* transmission between radials and cycling within the herd (Winfield and Groisman, 2003).

Some of the most commonly isolated *Salmonella* serotypes from swine have been Mbandaka (Funk et al., 2001), Derby (Davies et al., 1997), and Typhimurium (Letellier et al., 1999). In the current study, the most commonly isolated *Salmonella* serotype was Give (68.5%), followed by Mbandaka (18%) and Derby and Kentucky (3.7% each). It is to be noted that in our study, all of the Give and Mbandaka isolates were found in the wallows or in fresh feces near a wallow.

Coliform and generic *E. coli* bacteria are often regarded as sentinel marker organisms for fecal contamination in water supplies and the environment. However, *E. coli* have a low rate of survival outside animal hosts in the environment (Winfield and Groisman, 2003). Populations of coliform bacteria in the environment are considered indicative of the constant inflow of bacteria from fecal sources, yet coliforms are commonly found in the environment (Winfield and Groisman, 2003). The relatively constant generic *E. coli* populations found in the wallows in our study are indicative of frequent defecation within the wallows, suggesting a constant fecal-water/sediment-oral cycle.

Conversely, *Salmonella* spp. can survive in soil and sediments for over a year (Winfield and Groisman, 2003). The ability to survive passage through an external environment between host animals has been crucial for the evolution and success of *Salmonella* to be able colonize various diverse host species and be transmitted between nearby animals (Winfield and Groisman, 2003). Results from the current study support this idea, as genetically indistinguishable *Salmonella* serotypes were isolated from different wallows, as well as from the same wallow over more than 5 months. This result supports the data of Danish researchers that found a genetically indistinguishable *S.* Typhimurium was endemic to pig farms for a period of up to 2 yrs (Baloda et al., 2001; Sandvang et al., 2000). Therefore, our results indicate that wallows can be involved in a cycle of transmission and/or amplification of *Salmonella* within a *Salmonella*-infected swine herd

CONCLUSION

In the United States, swine producers utilize a variety of production systems including confinement systems as well as outdoor extensive systems. No differences in *Salmonella* populations between swine raised under traditional confinement conditions and outdoor intensive conditions were detected in our study. However, it appears that wallows can be a significant source of horizontal transmission of foodborne pathogenic bacteria among swine raised outdoors. Further research into the role of wallows in horizontal pathogen transmission is necessary before recommending increasing outdoor swine production.

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