Long term shedding of *Salmonella* Choleraesuis following experimental infection of very young piglets

Anderson RA¹, Harvey RB¹, Genovese KJ², Stanker LH³, DeLoach JR³, and Nisbet DJ¹

¹USDA/ARS Food Animal Protection Research Laboratory, College Station, TX 77845
²Texas A&M University, College Station, TX 77843
³MS BioScience Inc., Madison, WI 53704

Introduction

In the United States, more than 90% of diagnosed swine salmonellosis cases can be attributed to infections by *Salmonella* Choleraesuis, the etiologic agent of swine paratyphoid (9). Salmonellosis caused by this serotype is primarily manifested as a post weaning septicaemia or enterocolitis and often occurs on operations that commingle pigs of different ages (9). *Salmonella* Choleraesuis is rarely isolated from sources other than swine which suggests an important role of carrier swine in the spread of this host adapted pathogen (4-6, 9). Since *Salmonella* Choleraesuis infections can recur in pigs previously infected, an important role for latent carriers is further implicated (9). The development of a *Salmonella* Choleraesuis carrier state has been investigated experimentally with weaned piglets (1, 4-6) but not much is known regarding the potential of neonatal piglets to become long term carriers. This may be because neonatal piglets, while capable of becoming infected, rarely exhibit clinical salmonellosis (9). The objective of the present study was to experimentally infect suckling piglets with *Salmonella* Choleraesuis and then examine their daily shedding pattern and pattern of tissue colonization well beyond the initial infection period.

Materials and Methods

Twelve piglets were infected at two days of age via oral gavage as previously described (1) with 1.1 x 10⁶ colony forming units (CFU) of a novobiocin (NO) and nalidixic acid (NA) resistant strain. This mutant was propagated from a *Salmonella* Choleraesuis var. *kunzendorf* 3246pp isolate generously provided to us by Dr. Fedorka-Cray (Athens, GA, USA). The piglets were farrowed in a 1.5 m x 2.1 m farrowing crate (Hog Slat Inc., Newton Grove, NC, USA) and were weaned at 14 days of age to a concrete floored pen (approximately 6 m²). All piglets were reared in one pen until 54 days of age and were thereafter reared in two equal sized pens (six pigs per pen) except as noted below. Sixty-six days post challenge, two pigs from each pen were moved to the opposite pen so as to introduce a potential social stress. Eighty-four days post challenge, all pigs in one of the pens were subjected to potential transportation stress via a 10 hr round trip haul in an open top livestock trailer. The day of transit was sunny and temperatures ranged from 16 to 34°C (60 to 93°F). Pigs were phase fed starter and grower rations formulated to meet or exceed NRC requirements (8).

Because a wildtype salmonellae was isolated from the maternal dam 1 week prior to farrowing, rectal swabs collected in duplicate during the piglets first 23 days of age were cultured via 2 qualitative culture schemes which are described in detail elsewhere in these proceedings (2). This was done to ensure ample opportunity for recovery of wildtype salmonellae from our piglets yet also favoring recovery of our double antibiotic resistant challenge strain of *Salmonella* Choleraesuis. All other rectal swabs collected from these piglets (from 24 to 87 days of age) were cultured for the challenge strain only; however, specimens collected at necropsy were again cultured for both our challenge strain and for wildtype salmonellae (2). Since no other wildtype salmonellae were recovered from any of the above mentioned specimens, we present here only data pertaining to specimens cultured for recovery of the challenge strain.

Results

Shedding of *Salmonella* Choleraesuis was most frequent immediately post infection, with daily incidences (% culture positive) during the first 19 days post challenge ranging from 58 to 100% and a mean ± SD daily incidence of 80 ± 12% (Figure 1). The daily incidence of shedding declined thereafter, ranging from 0 to 58 % (mean ± SD daily incidence of 16 ± 13%) from days 20 to 85 post challenge. Subsequent commingling of selected pigs at 68 days of age had little effect on incidence of fecal shedding as did hauling of 6 of the pigs for 10 hr the day prior to necropsy (Figure 1). At necropsy (85 days post challenge), *Salmonella* Choleraesuis was recovered from cecal contents of 2 pigs, from spleen and tonsil specimens of 1 and 7 pigs, respectively, but not from colonic contents or either lung or ileocolic lymph node specimens of any of the pigs. Of these *Salmonella*-positive specimens, all except two of the tonsil specimens were obtained from the pigs that had not been transported. Throughout the study, only one pig (the one from which *Salmonella* Choleraesuis was recovered from splenic tissue) exhibited respiratory distress and then only after 66 days post challenge.
Discussion

Consistent with earlier reports regarding infection of weaned pigs (1, 4-6), we found that shedding of *Salmonella Choleraesuis* was most frequent immediately post infection (Figure 1). Earlier reports had suggested that various stresses, i.e., transportation, feed deprivation or commingling, may elicit increased shedding from animals (3, 7, 10-11) but neither commingling or transportation had much of an effect on shedding of *Salmonella Choleraesuis* by the pigs in our study. Also surprising is the fact that we did not recover *Salmonella Choleraesuis* from ileocolic lymph tissue collected at necropsy. Typically, these lymph specimens yield the best chance of recovering *Salmonella* from infected pigs (1, 4-6). Of the *Salmonella*-positive specimens, all except 2 were obtained from the non-transported pigs which is again contrary to our expectations. Hansen et al. (7) reported an increased colonic incidence of salmonellae in swine held in a slaughter facility's holding pens for 2 to 3 days before slaughter compared to swine slaughtered immediately upon arrival (35% versus 10% salmonellae incidence, respectively). Thus it may be that our relatively short post-transit sampling interval (<24 hr) did not allow enough time for proliferation of salmonellae to occur in response to the suspected transportation stress. Regardless, our findings reveal that the effect of various stresses on salmonellae shedding are not fully understood and that further research is needed in this area. Our results also show that *Salmonella Choleraesuis* can infect neonatal pigs and that these pigs may become long-term carriers capable of shedding the pathogen for up to 87 days of age.

![Figure 1. Daily incidence of *Salmonella Choleraesuis* shedding following experimental infection of 2 day old piglets.](image)

References


Acknowledgements

We thank Jim Snodgrass for his technical assistance. We thank Jackie Kotzur and Richard Burgess for animal care.

1999 ISECSP: Detection/Host-Agent Interaction 17