Competitive Exclusion of *Salmonella* from the Gut of Neonatal and Weaned Pigs

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**ABSTRACT**

Our laboratory has developed a bacterial competitive-exclusion (CE) culture against enteropathogens (which are considered human foodborne pathogens) for use in swine. In this article, we document the effects of this CE culture, PCF1, on cecal colonization by and fecal shedding of *Salmonella* Choleraesuis in neonatal and weaned pigs and its effects on the horizontal transmission of this pathogen between weaned penmates. Piglets treated with the PCF1 culture twice within their first day of life and challenged with *Salmonella* 48 h after birth shed *Salmonella* at a significantly (P < 0.05) lower rate than did control pigs in experiment 1. Significant reductions of the pathogen were also observed in the cecum, the cecal contents, the ileocolic junction, and the colon contents (P < 0.05). In experiment 2, culture of the cecal contents and lymph nodes revealed a significant reduction in *Salmonella* isolated from PCF1-treated pigs (P < 0.05). Pigs in experiment 3 were treated as pigs in experiments 1 and 2; however, they were followed through day 10 postweaning. Significant reductions in shedding were noted for treated groups both pre- and postweaning (P < 0.05). Experiments 4 and 5 assessed the effects of PCF1 treatment on the horizontal transmission of *Salmonella* between littermates that were followed through day 14 postweaning. In these experiments, litters were divided into untreated contacts (UC), untreated seeders (US), treated contacts (TC), and treated seeders (TS). Overall, TC in experiment 4 shed *Salmonella* at a significantly lower rate than UC and US did (P < 0.05). In experiment 5, the transmission of *Salmonella* was significantly reduced for litters in which TS or TC were present, as evidenced by reduced shedding of *Salmonella* by both treated and untreated animals within these litters (P < 0.05). TS shed less often than US did, resulting in reduced levels of *Salmonella* shedding by both treated and untreated contacts (P < 0.05). Litters containing both TC and UC or both TC and US also shed *Salmonella* at lower rates than did litters in which only UC and US were present (P < 0.05).

*Salmonella* has been isolated from nearly all vertebrate hosts from which it has been sought, with the possible exception of healthy fish in unpolluted water. Swine, cattle, and poultry are known carriers of salmonella (7, 31, 41). *Salmonella* has also been associated with foodborne illness in humans (7). Humans are typically infected with salmonellae through the ingestion of contaminated food or food products, and infection with salmonellae usually results in severe gastroenteritis (22). Transmission of the pathogen among swine can occur through the fecal-oral and intranasal routes, involving colonization of and dissemination from the gastrointestinal tract and organs such as lungs and tonsils, respectively (18).

*Salmonella* colonizes and inhabits the ceca of swine (14), and in poultry the cecum has been shown to be the primary site of salmonella colonization (28). The cecal environments for adult swine and poultry are similar in terms of pH, oxidation reduction potential, anaerobicity, and bacterial populations because swine and poultry are both hindgut fermenters and are fed very similar diets under commercial production conditions. The digestive tract of the newborn pig is usually sterile but rapidly develops a microflora characteristic of the species as the pig is exposed to a traditional commercial environment (23). Essentially sterile at the time of hatching, the intestinal tracts of poultry are rapidly colonized by microorganisms from the environment (29). The presence of a stable gastrointestinal microflora aids an animal in resisting infections, particularly in the gastrointestinal tract (26). This phenomenon has been referred to as bacterial antagonism (19), bacterial interference (16), the barrier effect (17), colonization resistance (49), and competitive exclusion (CE) (32).

The mechanism by which indigenous gut flora prevent salmonella colonization is not clear. Lloyd et al. (32) proposed that normal gut flora adhere to the intestinal-cecal epithelial cells and exclude salmonellae from essential microhabitats. Snoeyenbos et al. (43, 44) suggested that protection is due to direct competition for attachment sites. Soerjadi et al. (45, 46) reported that native gut flora adhere to the cecum as a mat of interconnected cells that may prevent the attachment of salmonellae, while Snoeyenbos et al. (44) suggested that in addition to adherence competition, normal gut flora metabolites may contribute to salmonella colonization control. Nisbet et al. (36) demonstrated that the oral administration of a CE culture rapidly established itself in the ceca of newly hatched chicks and...
resulted in a 100-fold increase in microbial populations for 3-day-old chicks compared with those for untreated controls. Additionally, this increase was highly correlated with increased concentrations of cecal total volatile fatty acids, especially propionic acid, and reductions in Salmonella Typhimurium cecal colonization levels. Furthermore, with the use of electron scanning microscopy it was shown that the CE bacteria preferentially colonized the crypts of the cecal mucosal epithelium, a primary site of salmonella colonization and invasion (15). The production of short-chain volatile fatty acids by anaerobic bacteria in the ceca was reported to inhibit Salmonella colonization in mice (9) and has been proposed to inhibit enteropathogens in poultry (5, 6) and swine (39, 40). The gastrointestinal volatile fatty acid profiles for pigs at weaning have been shown to decrease during the first 10 days postweaning, a period associated with enteropathogen colonization (33). The bacterial microflora in the mouse has been shown to undergo distinct changes as a result of weaning, and enteropathogen control in the gastrointestinal tracts of weaned mice has been associated with the concentration of butyric acid (32). Undissociated volatile fatty acids have also been reported to have an anti-enteropathogen effect in swine (40). Competition between normal flora and Salmonella for limited nutrients has also been proposed to be a mechanism that may control Salmonella growth (4–6, 26). In studies involving the use of continuous-flow (CF) cultures as models of the mouse intestinal ecosystem, Freret et al. (20, 21) and Wilson and Freret (50) proposed that the population dynamics of normal flora and invading enteropathogens may be regulated by competition for one or a few limiting nutrients.

Recently, with the use of microflora obtained from adult swine, our laboratory has employed a CF culture technique previously approved for use in the commercial poultry industry to develop a CE culture for swine (35, 37). Anderson et al. (3) have demonstrated that treating pigs with this CF culture of mixed species of bacteria present swine microflora (PCF1) decreases the concentration and incidence of Salmonella Cholaerasuis in the ceca of weaned pigs and decreases fecal shedding. In this report, we document the effects of PCF1 on cecal colonization by and fecal shedding of Salmonella Cholaerasuis in baby and weaned pigs and on the horizontal transmission of this pathogen among weaned penmates.

**MATERIALS AND METHODS**

**CF culture.** The porcine-derived CF culture (PCF1) was propagated from cecal contents collected from a 6-week-old healthy pig and was maintained through CF culture as described for avian cultures (12, 13). Briefly, cecal contents (ca. 50 g) were collected from the pig (which had been obtained from a commercial producer and maintained in our facility on a typical commercial diet that was free of antibiotics) and immediately transferred to an anaerobic chamber (Coy Laboratory Products, Ann Arbor, Mich.). The cecal contents were added to 100 ml of anaerobic Viande Levure broth medium (tryptose [10 g/liter], beef extract [2.4 g/liter], yeast extract [5 g/liter], dextrose [2.5 g/liter], and NaCl [2.5 g/liter]), and this mixture was immediately used as a seed culture for the CF culture system. For CF culture of the mixed cecal microflora, a BioFlo I fermenter fitted with a 2,000-ml chemostat vessel with an 1,150-ml working volume was used (New Brunswick Scientific Co., Edison, N.J.). The chemostat vessel containing 1,000 ml of Viande Levure broth was constantly flushed with a stream of O2-free CO2 to maintain anaerobic conditions. The medium was prepared in 13-liter Pyrex bottles, autoclaved for 1.5 h, and flushed with a constant stream of O2-free CO2 immediately upon removal from the autoclave. The chemostat vessel was filled with 1,000 ml of the Viande Levure medium (pH 5.5) and allowed to sit for 48 h before inoculation to ensure that there had been no microbial contamination prior to inoculation. The vessel was inoculated with the above-mentioned inoculum, the nutrient pump was turned on, and the culture was incubated under CF conditions. The dilution rate for the CF culture was 0.0446 per h. The CF culture was monitored daily for fermentation products and pH. After five vessel turnovers, a constant pH (6.0 to 6.2) was observed and the culture was deemed to be in a steady-state condition. This method of maintaining CF cultures has previously been used in our laboratory for the successful maintenance of CF cultures of poultry gut origin (42, 43). The PCF1 culture (38) contains at least seven of the following bacterial species: Enterococcus faecalis, Streptococcus bovis, Clostridium clostridiforme, C. symbiosum, C. ramosum, Bacteroides fragilis, B. distasonis, B. vulgaris, B. uniformis, and B. caccae. However, the culture is not limited to these species.

**Salmonella.** Salmonella used for experimental challenges was propagated from a primary pig isolate of Salmonella Cholaerasuis var. Kunzendorf, 3246. This isolate was selected on the basis of its resistance to both novobiocin (NO) and nalidixic acid (NA) in our laboratory and was maintained in tryptic soy broth medium containing NO at 25 μg/ml and NA at 20 μg/ml. All experimental Salmonella challenge materials were prepared from an overnight culture that had been serially cultured two consecutive times at 37°C for 24 h each. The challenge doses of Salmonella Cholaerasuis were determined on the basis of viable cell counts following overnight incubation at 37°C on brilliant green agar (BGA; Oxoid, Unipath Ltd., Basingstoke, Hampshire, UK) supplemented with NO at 25 μg/ml and NA at 20 μg/ml (BGA NO/NA).

**Animal experiments 1 and 2.** Pregnant sows were purchased from a commercial producer and maintained in our laboratories on a commercial diet that was free of antibiotics. Sows were cultured for the presence of wild-type salmonellae on arrival and up to the time of experimental challenge by previously described methods (30). With the exception of swabs from sows and baby pigs in experiment 1 (see “Results”), wild-type salmonellae were not detected in rectal swabs collected from sows prior to farrowing or from piglets for the 2 days immediately preceding experimental challenge. The prechallenge swabs were cultured via preenrichment in GN-Hajna broth (Difco Laboratories, Sparks, Md.), further enrichment in Rappaport-Vassiliadis broth (Difco), and selective differentiation on BGA plates containing NO (BGA NO plates) (27). Treated pigs were provided with a 5.0-ml oral dose of the PCF1 culture (109 CFU/ml of culture) from a sample that was withdrawn from the fermenter and transferred into sterile O2-free serum bottles. The culture was given to treated pigs within 30 min of withdrawal from the fermenter. Treatment was provided within 4 h of birth and again 24 h later. Piglets were challenged by intranasal inoculation with 2 ml of NO- and NA-resistant Salmonella Cholaerasuis (109 CFU/ml) 48 h after PCF1 treatment in experiment 1. Control piglets were challenged similarly; however, no PCF1 treatment was provided. One week postchallenge, piglets were euthanized by injection with sodium pentobarbital and necropsied for the collection of ileocolic lymph nodes, cecal contents, and, in some of the experiments, tonsils,
ileocolic junctions, livers, spleens, lungs, and colons. Rectal swabs, tissues, and cecal contents collected from piglets after challenge were incubated overnight at 37°C in GN-Hajna broth, transferred to Rappaport-Vassiliadis broth and incubated overnight at 37°C, and then streaked on BGANOBA culture for Salmonella Choleraesuis. Plates were examined for colonies exhibiting typical salmonella morphological characteristics, and suspect colonies were confirmed via serum agglutination with the use of Salmonella Antiserum Poly A IJV and Group C1, Factors 5 and 6 (Difco). Several representative colonies were also sent to the National Veterinary Services Laboratory (Ames, Iowa) for serotyping, and all colonies were confirmed to be Salmonella Choleraesuis var. Kunzendorf. In experiment 2, piglets were handled in a similar manner but were challenged via oral administration of 2 ml of NO- and NA-resistant Salmonella Choleraesuis (10^5 CFU/ml). Cecal contents were serially diluted in phosphate-buffered saline and were then spread plated on BGANOBA plates, and Salmonella counts were obtained. All animals were cared for according to standard swine husbandry practices and were fed (ad libitum) a typical commercial diet (corn and soy bases) formulated to be free of antibiotics and to meet or exceed nutrient requirements.

Animal experiments 3 and 4. Sows and piglets in experiments 3 and 4 were subjected to the same animal husbandry practices used in experiments 1 and 2. Sows and piglets were determined to be free of wild-type salmonellae prior to the experimental challenge. Treated piglets were provided the PCF1 culture within 4 h of farrowing and again 24 h later. In experiment 3, all piglets were orally challenged with 10^5 CFU of Salmonella Choleraesuis; on day 14, piglets were weaned from the sows and litters were housed in separate pens. Rectal swabs were taken daily from the day after experimental challenge until the termination of the experiment (10 days postweaning), and Salmonella incidences and cecal concentrations were determined as in experiments 1 and 2. In experiment 4, treated piglets were provided only one dose of the PCF1 culture, and this dose was provided within 4 h of birth. Experiment 4 was designed to measure the effects of horizontal transmission on the incidence of Salmonella shedding, which was determined with the use of daily rectal swabs. In the first litter of pigs, the piglets were divided into two groups, designated contacts and seeders. Seeder piglets were orally challenged with 10^5 CFU of Salmonella Choleraesuis 48 h after birth, and the remaining piglets were designated contact piglets and were unchallenged; neither group in litter 1 was provided PCF1. This experiment was carried out to determine the extent of horizontal transmission of the pathogen from infected to noninfected pigs. Rectal swabs were taken from piglets daily from the day they were born until the end of the experiment. Piglets were weaned at 14 days of age, and the litter was housed in a pen. Litters 2 and 3 were handled in the same manner except that these litters were divided into three different groups: seeders, untreated contacts, and PCF1-treated contacts. For these litters, PCF1 was provided to the treated contact piglets once within 4 h of birth, and the seeder piglets were challenged with Salmonella Choleraesuis 24 h later. This experimental design allowed us to determine the effect of PCF1 on the horizontal transmission of Salmonella Choleraesuis from the challenged seeder pigs to the untreated and treated contacts and was chosen because it best represents what most likely occurs under commercial conditions in the swine industry. For all three litters in experiment 4, ear tags were used to identify the individual piglets as seeders, untreated contacts, or treated contacts.

Animal experiment 5. Sows and piglets were subjected to the husbandry practices described above. Experiment 5 was conducted to assess the effects of a single dose of PCF1 within 4 h of the birth of pigs on the horizontal transmission of Salmonella Choleraesuis between littermates. Litters were as follows: litter 1 (n = 6)—seeders, untreated contacts; litter 2 (n = 8)—seeders, PCF1 contacts; litter 3 (n = 8)—PCF1 seeders, PCF1 contacts; litter 4 (n = 5)—PCF1 seeders, untreated contacts; litter 5 (n = 13)—seeders, untreated contacts, PCF-1 contacts; litter 6 (n = 10)—PCF1 seeders, untreated contacts, PCF1 contacts. Both untreated seeders and PCF1 seeders were orally administered 2 ml of Salmonella Choleraesuis (10^7 CFU/ml) 48 h after birth. Beginning the day after Salmonella challenge, rectal swabs were taken daily from each pig and examined for the presence of Salmonella Choleraesuis. Seeders, untreated contacts, treated seeders, and treated contacts were divided within the litters. Piglets were then handled as described in experiment 4.

RESULTS

Experiment 1. Analysis of rectal swabs indicated that sows were free of salmonellae when they first arrived at the facility. However, prior to farrowing, the control sow was shedding a serogroup B Salmonella. In addition, the piglets of both the control sow and the treated sow were also determined to be shedding a wild-type serogroup B Salmonella. No further identification of the serogroup B Salmonella was carried out. Piglets were challenged with the Salmonella Choleraesuis (serogroup C1) challenge organism (with the use of intranasal administration); therefore, the Salmonella incidence data obtained in experiment 1 represent total salmonellae and not just the Salmonella Choleraesuis challenge organism. The control piglets shed the serogroup B Salmonella exclusively, as determined by the analysis of rectal swabs, whereas in tissue samples there was a mixture of the challenge organism (Salmonella Choleraesuis) and the serogroup B Salmonella. For the PCF1-treated piglets, a mixture of the wild-type serogroup B Salmonella and the challenge Salmonella (Salmonella Choleraesuis) was found in the rectal swabs, but only Salmonella Choleraesuis was isolated from tissue samples. No consistent differences between the incidence of Salmonella-positive tissue samples for treated piglets and that for control piglets with respect to treatment were observed (data not shown). For the gut samples, total Salmonella incidences were significantly reduced (P < 0.05) in the ceca, in the cecal and colonic contents, and in the ileocolic junctions of treated piglets compared with those for control piglets (Table 1).

Experiment 2. Sows and piglets used in experiment 2 were determined to be free of wild-type salmonellae prior to experimental challenge. The Salmonella Choleraesuis challenges in this experiment and in the remaining experiments were administered orally, whereas in experiment 1 the challenge was administered via intranasal instillation. Treated piglets did not shed Salmonella Choleraesuis at any time during the experiment, as indicated by rectal swab
TABLE 1. Effects of porcine competitive-exclusion culture on incidence of gut colonization by, and fecal shedding of, Salmonella in suckling pigs*

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of pigs positive for Salmonella</th>
<th>No. of pigs tested (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control pigs</td>
<td>Treated pigs</td>
</tr>
<tr>
<td>Rectal swab</td>
<td>72/72 (100) A</td>
<td>10/56 (18) B</td>
</tr>
<tr>
<td>Cecum</td>
<td>9/9 (100) A</td>
<td>0/7 (0) B</td>
</tr>
<tr>
<td>Cecal contents</td>
<td>9/9 (100) A</td>
<td>2/7 (29) B</td>
</tr>
<tr>
<td>Ileocolic junction</td>
<td>9/9 (100) A</td>
<td>1/7 (14) B</td>
</tr>
<tr>
<td>Colonic contents</td>
<td>6/6 (67) A</td>
<td>0/7 (0) B</td>
</tr>
</tbody>
</table>

* Treated pigs were provided an oral dose of 5.0 ml of the porcine competitive-exclusion (PCF1) culture containing $10^9$ CFU/ml within 4 h of birth and again 24 h later. All treated pigs were intranasally challenged 48 h after the second PCF1 dose with $10^3$ CFU of Salmonella Cholerasuis. Control pigs were similarly challenged 72 h after birth. Values with different letters in the same row are significantly different ($P < 0.05$).

data. However, incidences of the shedding of total Salmonella were not significantly different ($P > 0.05$) between control piglets and treated piglets, because a low incidence of shedding was observed for the control piglet group (Table 2). Incidences of Salmonella Cholerasuis in ileocolic lymph nodes and cecal contents were significantly lower ($P < 0.05$) for treated piglets than for control piglets. In addition, Salmonella Cholerasuis counts for cecal contents were $>90\%$ lower for treated piglets than for control piglets ($P > 0.05$).

**Experiment 3.** Wild-type salmonellae were not isolated from either sows or piglets in experiment 3. Piglets in group 1 shed Salmonella at a significantly lower rate than did the challenge control pigs during the preweaning phase ($P < 0.05$) (Table 3). There was no difference in the shedding rate for control pigs and that for treated pigs in group 2 during the preweaning phase of the experiment. After weaning, both group 1 and group 2 pigs shed Salmonella at significantly lower rates ($P < 0.05$) than the controls did. The Salmonella count for the cecal contents of group 2 was $>2.5 \log_{10}$ units lower than that for those of control pigs, with none of the group 2 pigs culturing positive for Salmonella in either the cecal contents or the cecum itself. There was no difference in Salmonella counts for cecal contents for group 1 compared to controls.

**Experiment 4.** Wild-type Salmonella was not isolated from pigs during experiment 4. The numbers of positive rectal swab samples for treated contacts were significantly smaller than those for untreated contacts and seeders overall ($P < 0.05$) (Table 4). Numbers of positive samples for treated contacts were significantly smaller than those for seeders throughout the study ($P < 0.05$) (Table 4). However, although overall reductions in numbers of positive rectal swab samples for treated contacts were observed when all three litters in experiment 4 were compared, significant differences between the numbers for treated contacts and untreated contacts within litters 2 and 3 were not observed (Table 4). However, differences between total percentages of untreated and treated contacts were significant ($P < 0.05$).

**Experiment 5.** The results of experiment 5 are presented in Table 5. Sows and piglets were found to be free of wild-type Salmonella. Rates of Salmonella shedding for treated contacts were significantly lower than those for both treated seeders and untreated seeders, as indicated by analysis of daily rectal swabs ($P < 0.05$). Overall, treated seeders shed Salmonella significantly less often than did untreated seeders ($P < 0.05$). Treated contacts also shed Salmonella significantly less often than did untreated contacts when both treated and untreated contacts were present in a litter and when either group was present individually in a litter ($P < 0.05$).

**DISCUSSION**

The results of the present studies suggest that the use of a mixed-bacterial-species CE culture can reduce both the Salmonella count in the gut and the fecal shedding of these pathogens into the environment. Reductions in the fecal shedding of Salmonella resulted in diminished horizontal transmission between pen- and littermates. The reductions in Salmonella counts observed for PCF1-treated pigs and for pigs that came into contact with them may translate to less contamination in the slaughter plant and subsequent reductions in the contamination of pork products destined for human consumption.

Previous experiments conducted in our laboratory and elsewhere have demonstrated the efficacy of CE cultures (mixed- and single-strain cultures) in protecting swine against the enteropathogens Salmonella and Escherichia coli (3, 17, 24, 25, 47, 48) (see Blanco et al. (8) for E. coli virulence factors). Studies using single strains of Streptococcus spp. as microbial prophylactics against E. coli in

TABLE 2. Effects of porcine competitive-exclusion culture on incidence of gut colonization by Salmonella Cholerasuis in suckling pigs*

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of pigs positive for Salmonella</th>
<th>No. of pigs tested (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control pigs</td>
<td>Treated pigs</td>
</tr>
<tr>
<td>Rectal swab (incidence of shedding)</td>
<td>4/90 (4.4) A</td>
<td>0/72 (0) A</td>
</tr>
<tr>
<td>Rectal swab (pigs shedding Salmonella Cholerasuis at least once)</td>
<td>3/10 (30) A</td>
<td>0/1 (0) B</td>
</tr>
<tr>
<td>Ileocolic lymph nodes</td>
<td>10/10 (100) A</td>
<td>3/8 (38) B</td>
</tr>
<tr>
<td>Cecal contents</td>
<td>8/10 (80) A</td>
<td>3/8 (38) B</td>
</tr>
</tbody>
</table>

* Treated pigs were provided an oral dose of 5.0 ml of the porcine competitive-exclusion (PCF1) culture containing $10^9$ CFU/ml within 4 h of birth and again 24 h later. All treated pigs were intranasally challenged 48 h after the second PCF1 dose with $10^3$ CFU of Salmonella Cholerasuis. Control pigs were similarly challenged 72 h after birth. Values with different letters in the same row are significantly different ($P < 0.05$). Salmonella Cholerasuis counts for the cecal contents of control and treated pigs were significantly different at $2.9 \pm 1.97 \log_{10}$ CFU/g (range, 0 to 6.7 log$_{10}$ CFU/g) and $1.2 \pm 1.7 \log_{10}$ CFU/g (range, 0 to 3.8 log$_{10}$ CFU/g), respectively ($P < 0.05$).
swine were conducted in either gnotobiotic or caesarian-derived, colostrum-deprived pigs (47, 48). Experiments in our laboratory have involved the use of pigs farrowed and raised with the use of traditional swine production practices. The incorporation of traditional rearing practices into these studies allows the simulation of most, but not all, of the characteristics of a commercial swine system, and hence the results of these studies represent results that might be obtained if the studies were conducted on a commercial hog farm.

As in our work with CE in chickens, reduced horizontal transmission was observed for swine (11, 37). Anderson et al. (1, 2) found that the incidence of the transmission of Salmonella Choleraesuis by experimentally infected pigs was high (4 of 10 pigs exposed) when an oral dose of 10⁶ CFU was given to seeder pigs, but no transmission was observed for seeder pigs given lower doses. The present studies indicate that the administration of a dose of 10⁷ CFU of Salmonella Choleraesuis to seeder pigs was sufficient to result in transmission from seeder pigs to contacts. The rates of transmission of Salmonella were low for untreated contacts and greatly reduced for treated contacts (Tables 4 and 5). The apparent low incidence of the transmission of Salmonella Choleraesuis between swine in laboratory studies involving low doses of the bacteria makes seeder-contact studies difficult and results in the use of much higher doses of bacteria than are likely to be encountered in the production environment in order to enable researchers to measure differences in the rates of transmission between treated and untreated animals. Significant (P < 0.05) reductions in Salmonella counts for lymph nodes and cecal contents (Tables 1 and 2) were observed for treated pigs in experiments 1 and 2. In studies involving similar conditions and a challenge dose 10⁶ CFU of Salmonella Choleraesuis, Fedorka-Cray et al. (17) found similar reductions in the gut after piglets were administered a mucosal CE culture (MCES) soon after birth. Reductions in numbers of enteropathogens during the pre- and postweaning periods may translate to reductions in the overall contamination of pork products in the processing plant, although this possibility has not been investigated. Poultry studies suggest that reductions in Salmonella counts for young chickens translate to lower Salmonella counts for chickens prior to slaughter (10). Further studies will be conducted to investigate whether reductions in enteropathogens are seen at the finishing stage of production.

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of pigs positive/no. of pigs tested (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls (n = 6)</td>
<td></td>
</tr>
<tr>
<td>Rectal swab (fecal shedding preweaning)</td>
<td>26/57 (46) A</td>
</tr>
<tr>
<td>Rectal swab (fecal shedding postweaning)</td>
<td>61/102 (60) A</td>
</tr>
<tr>
<td>Cecal contents (incidence of Salmonella</td>
<td></td>
</tr>
<tr>
<td>Choleraesuis–positive pigs)</td>
<td>4/6 (67) A</td>
</tr>
</tbody>
</table>

* Treated pigs (groups 1 and 2) were provided an oral dose of 5.0 ml of the porcine competitive-exclusion (PCF1) culture containing 10⁶ CFU/ml within 4 h of birth and again 24 h later. All treated pigs were intranasally challenged 48 h after the second PCF1 dose with 10⁵ CFU of Salmonella Choleraesuis. Control pigs were similarly challenged 72 h after birth. Values with different letters in the same row are significantly different (P < 0.05). Salmonella Choleraesuis counts for the cecal contents of controls (2.81 ± 2.34 log₁₀ CFU/g; range, 0 to 5.8 log₁₀ CFU/g) and for the cecal contents of group 1 (1.65 ± 1.13 log₁₀ CFU/g; range, 0 to 3.5 log₁₀ CFU/g) were significantly different (P < 0.05) from the count for group 2 (0 ± 0 log₁₀ CFU/g).

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of pigs positive/no. of pigs tested (%)</th>
</tr>
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<tbody>
<tr>
<td>Group 1 (n = 8)</td>
<td></td>
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<tr>
<td>Rectal swab (fecal shedding preweaning)</td>
<td>8/63 (13) B</td>
</tr>
<tr>
<td>Rectal swab (fecal shedding postweaning)</td>
<td>74/148 (50) B</td>
</tr>
<tr>
<td>Cecal contents (incidence of Salmonella</td>
<td></td>
</tr>
<tr>
<td>Choleraesuis–positive pigs)</td>
<td>5/8 (63) A</td>
</tr>
</tbody>
</table>

* Treated pigs (groups 1 and 2) were provided an oral dose of 5.0 ml of the porcine competitive-exclusion (PCF1) culture containing 10⁶ CFU/ml within 4 h of birth. Untreated contact pigs were neither challenged with Salmonella Choleraesuis nor provided PCF1. Seeder pigs were challenged with 10⁷ CFU of Salmonella Choleraesuis. Piglets remained on sows until day 14 and were then weaned; individual litters were housed in separate pens until the termination of the experiment. Values with different letters in the same column are significantly different (P < 0.05).
swine production after the administration of the PCF1 culture to pigs as neonates.

REFERENCES


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**TABLE 5. Effects of porcine competitive-exclusion culture on horizontal transmission of Salmonella Choleraesuis between littermates**

<table>
<thead>
<tr>
<th>Group</th>
<th>Litter 1b</th>
<th>Litter 2b</th>
<th>Litter 3b</th>
<th>Litter 4b</th>
<th>Litter 5b</th>
<th>Litter 6b</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seeder</td>
<td>26/63 (41) A</td>
<td>28/63 (44) A</td>
<td>NP6b</td>
<td>40/72 (55) A</td>
<td>NP</td>
<td>94/198 (47) A</td>
<td></td>
</tr>
<tr>
<td>Treated seeders</td>
<td>NP</td>
<td>NP</td>
<td>20/84 (23) A</td>
<td>20/42 (47) B</td>
<td>NP</td>
<td>9/63 (14) A</td>
<td>49/189 (25) B</td>
</tr>
<tr>
<td>Treated contacts</td>
<td>NP</td>
<td>7/84 (8) B</td>
<td>2/84 (2) B</td>
<td>NP</td>
<td>0/84 (0) C</td>
<td>2/63 (3) B</td>
<td>11/315 (3) C</td>
</tr>
</tbody>
</table>

a Treated control pigs were provided an oral dose of 5.0 ml of the porcine competitive-exclusion (PCF1) culture containing 10⁶ CFU/ml within 4 h of birth. Untreated control pigs were neither challenged with Salmonella Choleraesuis nor provided PCF1. Seeder pigs were challenged with 10⁷ CFU of Salmonella Choleraesuis. Piglets remained on sows until day 14 and were then weaned; individual litters were housed in separate pens until the termination of the experiment. Values with different letters in the same column are significantly different (P < 0.05).

b NP, not performed.


