ORIGINAL RESEARCH

Cesarean section: A surgical method to derive pigs free of *Streptococcus suis*

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Summary

Purpose: *To determine whether* Streptococcus suis *of serotypes 1–34 colonizes the oropharyngeal region or the blood of pigs in utero.*

Methods: *Oropharyngeal and umbilical blood swab samples from 50 Cesarean-derived pigs, and oropharyngeal swab samples from their dams were collected. Pig samples were collected in a sterile bubble at the time of delivery before pigs came into contact with the sow or the environment. All samples were culturally examined for* S. suis. Streptococcus suis *isolates were serotyped.*

Results: Streptococcus suis *was not isolated from either oropharyngeal or umbilical blood samples from any pig.* Streptococcus suis *was confirmed in samples from 8 of 10 sows. The most prevalent serotype of* S. suis *isolated from sow samples was serotype 22 (62.5%) followed by serotype 9 (12.5%), serotype 23 (12.5%), serotype 4 (6.25%), and serotype 31 (6.25%).*

Implications: *The oropharyngeal region and blood of Cesarean-derived pigs of* S. suis *infected dams are free of those serotypes of* S. suis *isolated from these dams. Consequently, we hypothesize that Cesarean-derived pigs are more likely to be free of* S. suis *infection.*

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Streptococcus suis causes disease in pigs of all ages. Many serotypes of *S. suis* have been associated with a septicemia resulting in meningitis, arthritis, endocarditis, polyserositis, and pneumonia of pigs.[1] Streptococcosis is a bacterial disease that cannot be eliminated with the use of segregated (SEW) or medicated early weaning (MEW) procedures.[2–5] This resulted in greatly increased treatment and mortality costs in herds with streptococcosis problems.

Investigators have been attempting to determine the age at which a pig becomes infected with *S. suis* for many years. Clifton-Hadley, et al.,[6] concluded that *S. suis* may infect suckling pigs, but spread of the organism among weaned pigs in intensive production units was more frequent. The infection of suckling pigs with *S. suis* is becoming increasingly economically significant as early-weaning procedures become widely implemented in the swine industry. From the results of previous transmission studies,[4] we concluded that the sow sheds multiple serotypes of *S. suis* in bodily secretions and excretions. The pig can become colonized with *S. suis* shortly after birth when these sources are contacted. Pigs may also become infected with *S. suis* in utero or during birth. This hypothesis is supported by Robertson and Blackmore, who recovered *S. suis* from the reproductive tracts of slaughtered sows.[7] However, to develop effective prevention measures, the exact time *S. suis* is transmitted from dam to pig must be determined.

To understand this disease, an in-vivo model of streptococcosis in swine is needed. The model must include *S. suis*-free pigs and a method for inducing disease. Many researchers have developed protocols to induce streptococcosis in swine.[8–10] Clifton-Hadley did not detect *S. suis* type 2 from tonsils of 6- to 17-week-old hysterectomy-derived pigs maintained in quarantine.[11] It is not known whether these pigs carried other serotypes of *S. suis* or if a subclinical bacteremia existed in these pigs. The purpose of this study was to determine whether *S. suis* serotypes 1–34 colonize the oropharyngeal region or the blood of pigs in utero.

Materials and methods

Ten dams and 50 of their Cesarean-derived crossbred pigs from seven herds in Iowa were used. A 25% prevalence of *S. suis* among sows was assumed among the population. Thus, *S. suis* should have been detected in at least one sow with 95% confidence with the sample size of 10 sows. A sample size of five pigs per sow was used to detect *S. suis* with 95% confidence assuming the transmission rate from dam to pig was 50% (Episcope, K. Frankena and J.O. Goelema, Wageningen, the Netherlands) The adequacy of these sample sizes was confirmed in previous trials, in which we found *S. suis* colonization in seven of seven sows and 65 of 70 2-week-old pigs.[4,5]

Sample collection

All samples were taken using sterile swabs (S/P® Brand culturette system, Baxter Diagnostics Inc., Deerfield, Illinois.) The oropharyngeal region of each sow was sampled prior to surgery at day 113 or 114 of gestation. All pigs were derived by Cesarean section using a sterile bubble. Sterile swabs (removed from the original packaging) and surgical materials were placed in the bubble and resterilized with formaldehyde gas prior to each surgery. Cesarean sections were performed according to standard procedure.[12] The oropharyngeal region and blood from the umbilicus of each of five pigs per litter were sampled. All pig sampling occurred within the sterile bubble so that the delivered pigs had no sow or environmental contact.

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After these pigs had been transferred to a second sterile bubble, the oropharyngeal region of five additional pigs was sampled to control for the effects of formaldehyde sterilization on the culture swabs. The swabs used for sampling these pigs were sterile, but had not been resterilized with formaldehyde gas.

Cultural examination

Samples were plated onto sheep blood agar. After 24 hours of incubation at 36°C with 5% carbon dioxide, three 1- to 2-mm flat, α-hemolytic colonies were selected and streaked onto sheep blood agar for subculture. These isolates were then Gram stained and biochemically tested with catalase. Gram-positive, catalase-negative diplobacilli were considered to be *S. suis* suspects. Each suspect isolate was inoculated into a tube containing a 6.5% NaCl solution. Suspect isolates were also tested for acetoin and amylase production. The isolate was serotyped with antiserum to *S. suis* types 1–34 if there was:

- no growth in the 6.5% NaCl solution after 48 hours of incubation at 36°C, and
- no production of acetoin, but
- amylase was produced.

Serotyping was performed using a polyvalent coagglutination method followed by a monovalent coagglutination method. The culture was considered to be *S. suis* if it satisfied all of the above-mentioned criteria and agglutinated the antiserum. No conclusions were drawn from isolates that satisfied all requirements but did not agglutinate the antisera to *S. suis* 1–34 or that agglutinated antisera to all serotypes, even though these isolates could potentially be *S. suis* of other serotypes. We interpreted the identification of *S. suis* from any samples of pig origin to indicate failure of the Cesarean section method to obtain *S. suis*-free pigs.

Results

Pigs

Streptococcus suis was not isolated from either oropharyngeal or umbilical blood samples from any pig. Five samples (two oropharyngeal and three umbilical blood) from the pigs of one sow, and one sample (umbilical blood) from a pig of a second sow had 1–11 colonies per plate of a 1- to 2-mm, nonhemolytic, opaque, round, catalase-positive isolate, most likely *Staphylococcus* spp. There was no other growth on any plates.

Cultural examination of samples taken from the five additional pigs, using sterile swabs not exposed to formaldehyde, resulted in no growth of organisms.

Sows

Cultural examination of samples from all sows yielded 3–7 different colony types per plate. All plates contained small, round, alpha-hemolytic colonies, which were too numerous to count. *Streptococcus suis* was confirmed in samples from eight of 10 sows. Sixteen of the 30 total isolates from sow samples (three isolates per sow) were confirmed to be *S. suis* (Table 1). The most prevalent serotype of *S. suis* isolated was serotype 22 (62.5%), followed by serotype 9 (12.5%), serotype 23 (12.5%), serotype 4 (6.25%), and serotype 31 (6.25%). Six of the 30 isolates satisfied all biochemical criteria but did not agglutinate antisera to *S. suis* serotypes 1–34. Three of the 30 isolates satisfied all biochemical criteria and agglutinated antisera to all serotypes. Five of the 30 isolates were determined not to be *S. suis*.

Discussion and conclusions

Cultural examination of all samples demonstrated that the sows, but not the pigs, were infected with *S. suis*. Formaldehyde resterilization prior to use did not appear to affect the swabs since identical results were observed when swabs not exposed to formaldehyde were used for sampling. The *Staphylococcus* suspects isolated from six samples may have been due to contamination of the swabs during or after collection or the blood agar plates during inoculation. It is possible, but unlikely, that the pigs were carriers of this organism, considering the small number of isolates observed.

Our findings were consistent with those of others who could not isolate *S. suis* type 2 from the tonsils of Cesarean-derived pigs.[11] We have further determined that the oropharyngeal region and blood of Cesarean-derived pigs of *S. suis*-infected dams were free of all serotypes of *S. suis*, making the Cesarean section a proven method for producing pigs free from this organism. However, these results in conjunction with those of our previous transmission study did not support the

Table 1

Final diagnosis of isolates from sow samples

Sow ID (Farm of origin)	1 (A)	2 (B)	3 (C)	4 (C)	5 (D)	6 (E)	7 (F)	8 (F)	9 (F)	10 (G)	Total
Confirmed *S. Suis*	1	3	3	0	2	1	2	3	1	0	**16**
serotypes	9	4,22,22	9,22,22	—	22,22	23	22,22	23,22,21	22	—	
Unconfirmed *S. suis*	1	0	0	3	1	0	0	0	1	3	**9**
Not *S. suis*	1	0	0	0	0	2	1	0	1	0	**5**

Confirmed = Isolate satisfied all biochemical criteria and agglutinated antisera to one of *S. suis* serotypes 1–34
Unconfirmed = Isolate satisfied all biochemical criteria but did not agglutinate antisera to *S. suis* serotypes 1–34, or isolate satisfied all biochemical criteria but agglutinated antisera to multiple *S. suis* serotypes.

suggestion made by Clifton-Hadley and Alexander that hysterectomy and fostering was a successful method of populating a herd with *S. suis*-free stock.[11,13] Although the Cesarean-derived pigs were considered free of *S. suis*, these pigs would likely be infected by *S. suis* when they came into contact with the contaminated excretions and secretions of *S. suis*-infected nurse sows.[4] Cesarean-derived pigs may be used to populate nucleus herds if nurse sows are free of *S. suis* or if the pigs are artificially reared, andif strict biosecurity measures are taken to prevent introducing *S. suis* into the herd. Biosecurity measures should include eliminating rodents and flies.[14,15] Cesarean-derived pigs are suitable for use in research models, but may be impractical for commercial swine production of pigs free from *S. suis*.

Implications

- Cesarean-derived pigs are free of *S. suis* infection.
- Cesarean-derived pigs will be useful in research models for studying streptococcosis.
- Cesarean-derived pigs may be an impractical method to populate a commercial herd with *S. suis*-free pigs. However, these pigs will likely become infected with *S. suis* when they contact infected nurse sows, infected older pigs, or an *S. suis*-contaminated environment.
- It would be possible to commercially rear *S. suis*–free pigs if all-in–all-out management and *S. suis* free nurse sows are used. However, it would be difficult to maintain biosecurity and provide a *S. suis*-free environment for an extended period of time.

References

1. Sanford SE, Higgins R. Streptococcal diseases. In: Leman A, Straw B, Mengeling W, D'Allaire S, Taylor D, eds. *Diseases of Swine*. 7th ed. Ames, IA: Iowa State University Press; 1992:588–590.

2. Wiseman B, Morrison R, Dial G, et al. Medicated early weaning: Influence of age on growth performance and disease. *Proc Am Assoc Swine Pract.* 1992;469–475.

3. Clark LK, Hill MA, Kniffen TS, et al. An evaluation of components of medicated early weaning. *Swine Health and Prod*. 1994;2(3):5–11.

4. Amass SF, Clark LK, Wu CC. Source and timing of *Streptococcus suis* infection in neonatal pigs: Implications for early weaning procedures. *Swine Health and Prod*. 1995;3(5):189–193.

5. Amass SF, Wu CC, Clark LK. Evaluation of antibiotics for the elimination of the tonsillar carrier state of *Streptococcus suis*. *J Vet Diag Invest*.(In Press).

6. Clifton-Hadley FA, Alexander T, Enright MR. The epidemiology, diagnosis, treatment, and control of *Streptococcus suis* type-2 infection. *Proc Am Assoc Swine Pract*. 1986;473–491.

7. Robertson ID, Blackmore DK. Prevalence of *Streptococcus suis* types 1 and 2 in domestic pigs in Australia and New Zealand. *Vet Rec*. 1989;124:391–394.

8. Williams AE, Blakemore WF. Pathogenesis of meningitis caused by *Streptococcus suis* type 2. *J Infectious Diseases*. 1990;162:474–481.

9. Chengappa MM, Pace LW, Williams JA, et al. Efficacy of tiamulin against experimentally induced *Streptococcus suis* type-2 infection in swine. *JAVMA*. 1990;197:1467–1470.

10. Iglesias G, Trujano M. Experimental replication of clinical disease associated with *Streptococcus suis* type 2. *Agri-Practice.* 1992;13:25–30.

11. Clifton-Hadley FA, Alexander TJL. Studies of *Streptococcus suis* type 2 infection. *Pig Vet Soc (UK) Proc*. 1981;8:8–17.

12. Runnels LJ, Clark LK. Obstetrics. In: Leman A, Straw B, Mengeling W, D'Allaire S, Taylor D, eds. *Diseases of Swine*. 7th ed. Ames, IA: Iowa State University Press; 1992:588–590.

13. Alexander TJL. *Streptococcus suis*: An update. *Pig Vet Journal*. 1991;27:50–60.

14. Chanter N, Jones PW, Alexander TJL. Meningitis in pigs caused by *Streptococcus suis*— a speculative review. *Vet Microbiol*. 1993;36:39–55.

15. Enright MR, Alexander TJL, Clifton-Hadley FA. Role of houseflies (*musca domestica*) in the epidemiology of *Streptococcus suis* type 2. *Vet Rec*. 1987;121:132–133.