Streptococcus suis colonization of piglets during parturition

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Summary

Objective—To determine whether piglets were colonized with Streptococcus suis of sow origin during parturition

Materials and methods—Multiple samples were collected from 43 piglets of eight dams. Oral and vaginal swab samples were collected from each sow prior to farrowing. Piglets were removed from the vagina using individual sterile obstetrical sleeves into which they were immediately placed. Swab samples of the oropharynx and dorsal surface of each piglet were collected. Umbilical blood from each piglet was collected into tubes of culture media. Room air was sampled to estimate the concentration of airborne S. suis. All collected samples were culturally examined for S. suis. Streptococcus suis isolates were serotyped using antisera to S. suis serotypes 1/2–34.

Results—Fifty-four isolates of S. suis were identified. Multiple serotypes of S. suis were isolated from samples collected from a single animal in 11 of the 51 sows and piglets sampled. Streptococcus suis was isolated from the oropharyngeal samples of all of eight sows and from the vaginal swab samples from three of eight sows. Streptococcus suis was isolated from the oropharyngeal swab samples from nine of 43 piglets, the surface swab samples from 13 of 43 piglets, and the blood samples from two of 43 piglets. In three of eight dams, S. suis isolated from samples collected from the dam was of the same serotype as the S. suis isolated from oropharyngeal or surface swab samples of that dam's piglet. In two of these cases, the sow and her piglets matched on two different serotypes of S. suis. Streptococcus suis was not isolated from air samples.

Implications—In these studies, we concluded that the source of S. suis was the sow, and that S. suis was transferred to the dorsal surface and oral cavity of the piglet during parturition when the piglet came into contact with S. suis from sow vaginal secretions.

Keywords: swine, Streptococcus suis, serotypes, vertical transmission

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treptococcus suis infection in swine is a problem encountered by producers and veterinarians worldwide. Streptococcosis primarily affects nursery pigs, and cannot be prevented by either segregated or medicated early weaning (SEW, MEW) protocols. And serotypes of S. suis have been associated with a septicemia resulting in meningitis, arthritis, endocarditis, polyserositis, and

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pneumonia, primarily of nursery pigs. Streptococcus suis serotype 2 is widely considered the most common cause of clinical streptococcosis in swine, and is the focus of much research; however, in the herds we sampled in Indiana, other serotypes of *S. suis* are more commonly isolated from cases of meningitis. The production of a naive subpopulation of SEW pigs may render streptococcal virulence factors more important than serotype.

The age at which a piglet becomes infected with *S. suis* has been investigated for many years. Clifton-Hadley, et al., 6 concluded that *S. suis* type 2 may infect suckling piglets, but that the spread of the organism among weaned pigs in intensive production units was what was of prime importance. The importance of *S. suis* colonization of suckling piglets, however, is gaining increasing recognition as early weaning procedures become widely implemented in the swine industry. Piglets colonized at an early age carry *S. suis* into the nursery, where it may be transmitted among other weaned pigs and cause clinical disease as maternal immunity declines. From the results of previous transmission studies, 4 we concluded that the sow sheds multiple serotypes of *S. suis* in bodily secretions and excretions. Piglets can become colonized with *S. suis* shortly after birth when they come into contact with these sources.

Cesarean section is one method to produce piglets free from *S. suis*. Cesarean-derived piglets from *S. suis*-infected dams have been reported to be free of all serotypes of *S. suis*.^{7,8} However, cesarean section is not likely to be adopted as a method for obtaining piglets for commercial pork producers. Decluzeau, et al., have produced piglets free from certain organisms by surface decontamination of the piglets immediately after birth. Thus, surface contamination by *S. suis* of piglets during birth might be negated, if it occurs, and allow the production of nonsurgically derived piglets free of *S. suis* infection.

The purpose of this study was to determine whether *S. suis* colonized the oropharynx, surface, or blood of piglets during birth.

Materials and methods

Experimental design

The herd of origin was a 1200-sow, farrow-to-feeder-pig unit. *Strepto-coccus suis* serotypes 2, 3, 4, 5, 6, 7, 8, 9, 21, and 22 were known to exist in the herd prior to the study. ^{4,5} Clinical streptococcosis was not a significant problem in this herd. The study sample consisted of eight parity-two to parity-five crossbred sows from one weekly farrowing group, and their 43 newborn piglets, so that:

• six piglets were sampled from each of five sows (sows 1,2,3,5, and 6),

- five piglets were sampled from one sow (sow 8), and
- four piglets were sampled from each of two sows (sows 4,7).

A prevalence of at least 25% of *S. suis* was assumed among the population of 60 sows from one weekly farrowing group. Thus, the organism should have been detected in at least one sow with 90%–95% confidence with a sample size of eight sows. A 30% prevalence of *S. suis* was assumed among the population of piglets in a single litter (born alive = 10 piglets). Thus, the organism should have been detected in at least one piglet per litter with 90%–99% confidence if four to six piglets per litter were sampled (Epi Info version 5.01-b, 1991. Centers for Disease Control, Atlanta, Georgia). The adequacy of these sample sizes was confirmed in previous trials, in which we found *S. suis* colonization in all of seven sows and 65 of 70 2-week-old piglets. ^{4,5}

Sample collection

Farrowing was induced with 263 µg cloprostenol sodium injected intramuscularly (Estrumate[®], Miles, Shawnee, Kansas) 30 hours before the start of sample collection. Prior to farrowing, oral and vaginal swab samples were collected from each sow using sterile swabs (S/P[®] Brand culturette system, Baxter Diagnostics Inc., Deerfield, Illinois).

Using a sterile obstetrical sleeve, the piglet was removed from the vagina, and the sleeve immediately was inverted over the piglet and sealed. Sterile gauze was used to remove any fetal membranes covering each piglet's rostral surface. The oropharyngeal region and dorsal surface of each piglet (before it was cleaned) were swabbed with sterile swabs (S/P® Brand). The umbilical cord of each piglet was exposed and the distal ½ to ½ was removed using a sterile scissors. Approximately 1 mL of umbilical blood was collected by milking the blood into tubes of Todd-Hewitt broth (THB).

Room air was sampled at a rate of 1 cubic foot per minute onto sheep blood agar plates using an impactor (Gast Model 0523–101Q-G5820X, Benton Harbor, Michigan) for 5, 10, 15, and 30 seconds.

Cultural examination

Sow oral swab samples were plated onto sheep blood agar and incubated within 24 hours of collection.

Sow vaginal and piglet oral and surface swab samples were plated onto sheep blood agar within 11 hours of collection. The sow vaginal, piglet oral, and piglet surface swab samples were then enriched in THB for 12 hours, and a sample of the THB plated onto phenylethyl alcohol (PEA) agar with 5% sheep blood, and incubated for 18 hours.

Piglet umbilical blood samples were incubated within 11 hours of being collected and plated onto PEA agar after 12 hours of incubation in THB.

After 18–24 hours of incubation at 37°C with 5% CO₂, we selected up to three 0.5- to 1-mm flat, α -hemolytic colonies per site for each sow and piglet sampled. These isolates were then Gram stained and biochemically tested.

Of the Gram-positive diplobacilli that did not grow in 6.5% NaCl solution, a culture had to meet all of the following criteria to be considered

a S. suis suspect:

- catalase-negative,
- · acetoin-negative, and
- · amylase-positive.

Serotyping

All *S. suis* suspects were serotyped using a polyvalent coagglutination method and then a monovalent coagglutination method¹⁰ with antisera to *S. suis* types 1/2–34 (antisera provided by R. Higgins, University of Montreal, Saint-Hyacinthe, Quebec, Canada). Suspect isolates were considered to be *S. suis* if they agglutinated the antiserum. No conclusions were drawn from suspects that did not agglutinate the antisera to *S. suis* 1/2–34, even though these isolates could potentially have been *S. suis* of other serotypes.

Evaluation of results

The number of viable bacteria in the air samples was calculated using previously published methods.¹¹ Identifying *S. suis* from any samples of piglet origin was considered to be the result of colonization of that piglet during birth.

Results

Eleven serotypes of *S. suis* were identified in the fifty-four isolates:

- serotype 5 (50% prevalence),
- serotype 12 (14.8%),
- serotypes 10 and 34 (7.4% each),
- serotype 9 (5.6%),
- serotypes 8 and 27 (3.7% each), and
- serotypes 4, 11, 13, and 30 (1.85% each).

Multiple serotypes of *S. suis* were isolated from samples collected from a single animal in 11 of the 51 sows and piglets sampled. *Streptococcus suis* was isolated from the oropharyngeal samples from eight of eight sows and from the vaginal swab samples of three of eight sows (Table 1). In the litters of sows 4 and 5, no *S. suis* was isolated from any of the piglets. In the litter of sow 7, *S. suis* was isolated from all the piglets. In the remaining litters, *S. suis* was isolated from some but not all of the piglets.

A single serotype of *S. suis* was isolated from littermates in two of eight litters (Table 1). Multiple serotypes of *S. suis* were isolated from littermates in four of eight litters. *Streptococcus suis* was isolated from the oropharyngeal swab samples from nine of 43 piglets, the surface swab samples from 13 of 43 piglets, and the blood samples from two of 43 piglets. In three of eight dams, *S. suis* of the same serotype isolated from samples collected from the dam were detected in oropharyngeal or surface swab samples of that dam's piglet. In two of these cases, the sow and her piglets matched on two different serotypes. There were 8–16 colony-forming units of organisms per liter of room air; however, *S. suis* was not isolated from air samples.

Serotypes of *S. suis* isolated from each collection site

Sows			Piglets			
Sow ID	Oropharyngeal region	Vaginal tract	Piglet ID	Oropharyngeal region	Dorsal surface	Blood
1	5, 34	_	P1	_	_	
	Six piglets sampled		P2	_	_	_
			P3	11, 12	_	_
			P4	12	_	_
			P5	_	_	_
			P6		30	
2	8, 27	5, 12	P1	_	12	_
	Six piglets sampled		P2	_	12	_
	, 3	•	P3	_	_	_
			P4	_	5	_
			P5	_	_	_
	0.44		P6			
3	9, 12	5	P1	_	5	_
	Six piglets s	ampled	P2	_	5	_
			P3	12	5	_
			P4	5	5	_
			P5	5	5	_
	10.24		P6			
4	10, 34	_	P1	_	_	_
	Four piglets	sampled	P2 P3	_	_	_
			P4	_	_	_
5	9, 34		P1			
Ü			P2	_	_	_
	Six piglets s	ampieu	P3	_	_	_
			P4	_	_	_
			P5	_	_	_
			P6	_	_	_
6	10	_	P1	_	5	_
	Six piglets s	ampled	P2	_	_	_
	p.g		P3	_	_	_
			P4	_	5	_
			P5	_	_	_
			P6	_		_
7	4, 27	5 , 8	P1	5	_	_
	Four piglets	sampled	P2	5	_	_
	. car pigicis	campiou	P3	_	5	_
			P4	_	5	
8	34	_	P1	_	_	_
	Five piglets	sampled	P2	5, 10	_	_
			P3	5	_	_
			P4	_	_	5
			P5	_	_	13

Boldface serotype numbers and white backgrounds highlight instances where the same serotype was isolated in both a piglet and its dam.

Discussion

We previously reported that cesarean-derived piglets were free of *S. suis*, but 1-day-old piglets were colonized by *S. suis*. ^{4,8} Whether piglets were colonized with *S. suis* during parturition was not known.

These results indicated that the oropharyngeal area and vaginal secretions of sows were colonized by multiple serotypes of S. suis. However, S. suis was not isolated from piglets in two of eight litters or from the vaginal tracts of five of eight sows. Thus, either S. suis was not present at these sites or cultural examination did not detect the S. suis that was present. It is possible that the vaginal tract of the sow was not colonized or colonized in such low numbers that transmission did not occur from sow to piglet. Because we only chose three suspect isolates from each sample we collected, however, our ability to detect S. suis was limited. It is possible that S. suis was present at the sampled site, but not cultured, or that S. suis was cultured, but those isolates were not included in our sample of three suspects. Additionally, S. suis might have been present, but of a serotype other than 1/2-34, and consequently not detected. Serotype differences among sows and their piglets may also be explained by the low sensitivity of the culture procedure. Thus, piglets and sows may have been colonized by multiple serotypes of S. suis, but we were unable to detect all of them.

There are two possible explanations for the isolation of *S. suis* from the umbilical blood of two piglets:

- the sample might have been contaminated during collection because the blood was milked from the umbilical vessels.
- *Streptococcus suis* might have been drawn up the cord via the blood during the birth of the piglet when the end of the umbilicus contacted *S. suis*-infected sow vaginal secretions.

Our results provide strong evidence that in most cases, vaginal secretions from the dam were the initial source of *S. suis* that colonized the oropharynx and surface of neonatal piglets during parturition. The surface of the piglet appeared to be colonized as the piglet passed through the vaginal canal. The most likely explanation for the colonization of the oropharynx of piglets with *S. suis* was that they swallowed the colonized vaginal secretions of the sows during the birth process.

Our results are supported by Robertson and Blackmore, ¹² who reported that piglets derived from dams whose vaginas were colonized with *S. suis* type 2 became nasal carriers of *S. suis* type 2. We are currently analyzing the genomic DNA from the *S. suis* isolates of both sow and piglet origin in this study to further demonstrate vertical transmission at the molecular level.

It is possible that the samples were contaminated by airborne *S. suis*. This is unlikely, however, considering that *S. suis* was not isolated from the collected air samples, but large numbers of *S. suis* were isolated from pig and dam samples.

Surface disinfection of piglets at birth thus appears not to be a feasible method to prevent *S. suis* colonization of swine because in our study some piglets appear to have swallowed *S. suis* from vaginal secretions of the sow during the birth process, and in some piglets *S. suis* entered the navel. We hypothesize that the sow is colonized by multiple serotypes of *S. suis* and provides passive immunity to the piglet against these serotypes. The piglet is then colonized by some, none, or all of these serotypes of sow origin. As passive immunity declines, the pig becomes susceptible to those serotypes of *S. suis* that are not its "normal" flora. The pig is most susceptible to clinical streptococcosis at the time when it is in the SEW nursery and is exposed to multiple serotypes of *S. suis* being shed by other nursery pigs. Currently, cesarean derivation is the only method of obtaining *S. suis-*free piglets.

Implications

- Piglets are colonized with *S. suis* during the birth process when they contact and/or swallow *S. suis* from vaginal secretions of their dam.
- Surface decontamination cannot be relied upon to obtain *S. suis*-free piglets.
- Cesarean derivation is the only method of obtaining *S. suis* free piglets.

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