COMMENTARY

AI—How much of a PRRS threat?

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In the 7 years since the first recognized clinical outbreaks of PRRS virus, we have learned a great deal about this pathogen. However, many questions still remain unanswered, and the economic importance of the syndrome adds urgency to the pursuit of answers. One important question concerns the risk of transmitting PRRS through artificial insemination (AI). The use of AI in the swine industry continues to increase. Swine producers who have PRRS-negative herds are anxious about the possibility of introducing the PRRS virus (PRRSV) through AI.

Genetics must be added to a herd in some form. In terms of biosecurity, should a producer:

- avoid AI altogether;
- obtain semen only from PRRS-tested studs; or
- embrace AI as a safer method of genetic introduction as compared to the alternative of bringing in young adult breeding animals?

The answer to this question depends on two factors: the biosecurity level of the individual producer and the likelihood of transmitting PRRS via AI. Swine practitioners realize that the commitment to biosecurity varies widely in the industry. At one extreme there are the few isolated nucleus herds representing the top tier of breeding/health pyramids. Such farms might want to avoid AI altogether, or at least obtain semen only from PRRS-tested studs. At the opposite extreme are the numerous commercial herds located in hog-dense regions. Some of these units have purchased breeding stock from multiple sources for years. Moreover, some have never observed an appropriate isolation protocol for incoming breeding animals. When farms like this contemplate a move to AI, it may be imprudent to discourage them without a very good reason. After all, AI technology seems to circumvent many important swine pathogens as compared to the alternative of introducing breeding stock.[1] Is PRRSV an exception in this respect? Although at this time we can not definitively answer whether PRRSV can be transmitted via AI, pracitioners can provide some leadership after reflecting on the history and biology of the virus and after considering the available epidemiologic and experiemental studies.

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Historical perspective

Retrospective serologic studies on three Iowa swine serum banks from before, during, and after the onset of PRRS in that state indicate that PRRSV hit a naive swine population around 1985.[2] It is probably safe to assume that the entire North American swine population was similarly naive to this virus around 1980. Today the number of seropositive herds in the United States is estimated at 50%.[3] Even before the virus was discovered, the rapid spread of the disease suggested a viral etiology with respiratory and/or enteric transmission potential. This in fact turned out to be the case. Today it is accepted that pig movements and airborne spread have been the major transmission routes for the spread of PRRS among herds.[3] The fact that most of the early transmission experiments were conducted by intranasal inoculation of the negative test animals[4-6] and that the best cell line for isolation of the virus itself is the porcine alveolar macrophage (PAM)[7] underscore the likelihood that, in terms of transmission, PRRS is primarily a respiratory virus. Fecal shedding of the virus has also been reported.[8]

Alternative transmission routes for PRRSV, including the potential for transmission through semen, are less well established. This situation is not unprecedented. In considering pseudorabies virus (PRV) or swine influenza virus (SIV), we have examples of primarily respiratory contagions that happen to cause reproductive damage and yet are rarely, if ever, transmitted via AI. It seems plausible that PRRSV, like other viruses adapted for respiratory transmission, would have lower shed in semen and face a less-receptive route of entry in the female reproductive tract than in the respiratory tree. In the rare instance when a primarily respiratory virus does transmit venereally, it probably occurs during natural service within a herd that is experiencing an epidemic outbreak of the disease.

Biologic considerations

One of the main factors leading to the speculation about the potential for PRRS transmission via semen stems from the biological characteristics of several similar positive-stranded RNA viruses. Three of these non-arthropod Togaviruses appear to closely resemble PRRSV.[9] They include a mouse virus (lactate dehydrogenase-elevating virus—LDV), a monkey virus (simian hemorrhagic fever virus — SHFV), and an equine virus (equine arteritis virus — EAV). The mode of transmission of all three viruses primarily involves the nasopharygeal route by way of saliva (LDV) or respiratory droplets (SHFV and EAV).[9] Of the

three, EAV can also persist up to 2 years in the testes of some stallions and infection can spread to mares via semen, raising the possibility that PRRSV might behave in a similar fashion. Equine arteritis virus, however, is unique in this respect compared to the other two nonswine viruses in the group. Several other factors suggest that EAV may likewise differ from PRRSV.

One of the biological properties the nonswine viruses reportedly share is a predilection for macrophage cells.[9] Lactate dehydrogenase-elevating virus is the most restrictive of the three in that it can only be isolated using a cell culture of mouse peritoneal macrophages.[3] PRRSV, which is genomically most similar to LDV,[3] is best isolated on PAMs, but can also be isolated on CL 2621, a proprietary cell line.[7,10,11] In contrast, EAV readily replicates on a number of established cell lines in vitro.[12] One might speculate that EAV's less restrictive requirements for in vitro isolation/replication may have something to do with its in vivo ability to persistently infect the equine testes. In contrast, the PRRS virus's more restrictive nature, like that of LDV, likewise might preclude persistent testicular multiplication.

The second common characteristic of the three nonswine Togaviruses is that they establish persistent infections in the individual host animal. In swine, this characteristic would preclude the possibility of eliminating the disease from a herd by any means short of depopulation. This is not the case. Recently the virus was reported to spontaneously disappear from a previously infected herd.[13] Other studies have shown that nursery depopulation can break the cycle of infection on farrow-to-finish farms.[14] To date the evidence suggests that PRRSV rarely, if ever, results in persistent infection, testicular or otherwise, in individual animals.

In a recent bioassay,[15] experimentally infected boars were shown to have virus in their semen for up to 43 days. Although this is longer than the relatively short viremic period (9–14 days) in adult animals, it doesn't exceed the prolonged post-infection shedding period for the virus that has been described in some animals, apparently in the absence of viremia.[16,17] The presence of PRRSV in semen 43 days PI might well be expected considering that a one-time septicemic event could possibly affect the presence of virus in the next 50–60 days of sperm production — the time it takes a type-A spermatogonium to mature to ejaculated spermatozoa in the boar.[18] Even given the potential that PRRSV may be present in semen over a 1.5- to 2-month time frame, this is an order of magnitude different from the timeframe of EAV, in which persistently infected stallions have been reported to shed virus in their semen for up to 2 years. Moreover, the presence of virus in the semen, though necessary, does not obviate other necessary factors (e.g., the presence of alveolar macrophages) that may be lacking for the successful transmission of the virus via AI. It is noteworthy that in a study of six AI centers (230 boars per center) that became infected with PRRS, Feitsma, et al.,[19] reported that the semen collected from affected boars did not infect the sow herds in which it was used.

Epidemiologic evidence

At the time when the clinical PRRS epidemic in the United States was at its height, the swine AI industry was by today's standards rather small. Today with AI technology being implemented on more and more farms, the incidence of newly infected PRRS herds is declining. It is interesting to note this inverse relationship, but no causality or lack of it can really be inferred. As pointed out in a recent review,[3] we are witnessing a new respiratory contagion settling into a now not-so-naive swine population. But if AI had played even a minor role in the farm-to-farm dissemination of the disease during the epidemic period, it seems likely that there would be more than a few circumstantial reports implicating that route.

A case in point is the Yeager, et al., study[20] from South Dakota. Although a outbreak occurred 2 weeks after semen was introduced from a boar stud of unknown status, the report mentions that the involved herd was a "large" 200- to 225-sow farrow-to-finish operation, and that it was within 3 km of the nearest neighboring swine farm. The report does not identify the PRRS status of the neighboring swine farm, so presumably it was not determined as part of the investigation. However, according to British case-control studies[21] done to assess risk factors for PRRS transmission during that country's PRRS outbreak, the infected South Dakota herd was at greatest risk from both its size and its proximity to the neighboring herd. Airborne spread over distances of 3–5 km has been well documented.[21,22]

Because the most convincing epidemiologic evidence for PRRS transmission via AI comes from Great Britain,[21] it seems worthwhile to closely review that situation. The situation occurred at the height of the PRRS epidemic in Britain, when the virus was, for a time, a reportable disease. Although the published report indicates that several boar studs were involved, I am aware of hitherto unpublished confounding factors associated with at least one of the implicated studs.[23] In February 1992, this AI center's 40-boar stud in the north of England experienced a transient illness in about 10 boars. The local attending veterinarian submitted blood for PRRS serology as part of the workup. Some of the samples were positive, so the regulatory officials became involved and did a whole-herd test and found that 85% of the stud had seroconverted. The stud was subsequently shut down for 10 weeks under statutory regulations.

During that time, the Ministry of Agriculture conducted a traceback to determine whether semen from the center had precipitated infection in recipient herds. The Ministry then checked with 1200 customers receiving semen during the period the stud was thought to have been in the acute stages of PRRS infection, and discovered only four herd outbreaks that couldn't be explained by animal movement or airborne transmission. Interestingly, the stud never experienced any decrease

in semen production, although the Fietsma, et al.,[19] study indicates it would have been expected if a virulent strain of the virus had caused the illness. As a result, the AI center's supervising veterinarian[23] became suspicious of the original diagnosis, and had the original serum samples tested for SIV. (Incidentally, England was at that time in the midst of an epidemic of a new strain of SIV in conjunction with the PRRS epidemic.[24]) All of the sera were positive for SIV.

In their published report implicating semen, the Ministry of Agriculture[21] stated, "All eight herds had used artificial insemination with semen from herds suspected (retrospectively) to be in the acute stages of PRRS infection at the time of semen collection." Given the serologic evidence that another virus (i.e., SIV) may have produced the original transient illness in one of the studs, a fact in part supported by the absence of negative effect on sperm production, the ministry's assumption about this one stud's PRRS infectivity must at least be questioned. Indeed, the Ministry of Agriculture report does mention that "early in the epidemic, finding PRRS antibodies in acute sera was taken as sufficient evidence to confirm an outbreak," and that later "as the infection has spread it has become more important to confirm active infection by demonstrating seroconversion in paired samples." The report indicates that attempting to identify infected boar studs was confusing because of the "apparent increase in incidence of swine influenza associated with a new variant of H1N1 virus." So, it seems that in at least four of the eight herds that initially were believed to have been infected via AI, the boar stud's infectivity could reasonably be called into question. Even without the possible confounding influence of SIV, the recipient herds supplied by that one stud had only a 0.3% (4 in 1200) incidence of infection. In a recent review article, the author sums up the epidemiologic evidence as follows, "Field observations indicate that transmission (via semen under natural conditions), if it occurs, is possible for a short time (< 1 week) only."[3]

Semen transmission experiments

To date, three experimental studies seeking to transmit PRRSV using either unextended or extended semen have been published. In the first study,[15] Swenson, et al., reported that unextended semen from recently infected boars (up to 43 days post-infection) could produce infection in negative test animals when the semen was injected intraperitoneally. In the second study,[20] Yeager, et al., found that semen collected from a recently infected boar and infused directly (i.e., not extended) into the uterus of negative test gilts produced infection in the gilts. The third study[25] used extended semen from a recently infected boar and the researchers did not observe an infection when the extended semen was artificially inseminated into the test gilts. However, unextended semen from the same boar did produce infection in young pigs if injected intraperitoneally. It is noteworthy that the use of unextended versus extended semen interperitoneally in the third study does not rule out the possibility that the process of extension may somehow destroy the virus. While this third study should be replicated, the initial finding reminds us that other factors, such as route of entry and virus quantity, affect the outcome of exposure and not just the presence of viable pathogen.

The preliminary conclusions one can draw from the laboratory transmission experiments are:

- recently infected boars can shed virus in their semen for at least 43 days post-infection;
- unextended or "raw" semen from recently (6 days post-infection) infected boars can produce infection if infused into the reproductive tract of susceptible gilts;
- the reproductive tract of susceptible pigs may offer some barrier to infection as compared to the intraperitoneal environment;
- the process of extending semen may or may not destroy the virus, but it probably at least decreases the infectivity of the semen by simple dilution.

Taken together, this research seems to indicate that like other viral infections, the transmission of PRRS is related to the dose of the virus and the way it enters the body. In particular the research on extended semen seems to show that, for PRRSV, the intrauterine route is not particularly receptive to infection and that use of extended semen at least reduces the risk of experimental infection by this route. Though the Yeager, et al., study[20] demonstrated that a 60-mL volume of unextended semen from recently infected boars could be used to infect gilts by the reproductive route, the author still has no knowledge of any swine that have been experimentally infected with PRRS using extended semen in AI.

Conclusions

Although studies to date, both experimental and epidemiological, have not made a strong case for the importance of AI in the transmission of PRRSV from farm to farm, further research may prove the possibility more conclusively. Certainly more research is warranted to clarify the risk. In the mean time, and given that the biosecurity needs on some farms may dictate to the contrary, practitioners should probably be reluctant to discourage the use of AI on most swine farms simply on the basis of PRRS-phobia. The incidence of new cases of the disease has slowed. We're no longer in the midst of a national epidemic of the disease, and boar studs are developing PRRS programs to test and monitor themselves to ensure a PRRS-safe product. Also, boar studs are relatively small populations, and thus less susceptible to an airborne PRRS strike than are the larger farrow-to-finish seedstock operations.[21] Even in the event of a subclinical PRRS outbreak, the virus would be expected to burn itself out more quickly in an adult boar population than in a conventional farrow-to-finish seedstock

enterprise where a more protracted course of infection is typical.[26] It follows, then, that contaminated genetic product could be unknowingly sold for a longer period of time from a PRRS-infected seedstock herd than from a PRRS-infected AI center. Finally, the evidence is persuasive that moving young adult breeding animals is far riskier than moving semen.[21]

The commercial swine industry is replete with case histories about disease outbreaks after breeding animals have been introduced. PRRS is currently a popular disease and an important one, but in the end it is just one of a basketful of swine pathogens that can undo production on hog farms. One of the main reasons for the explosive growth of AI in swine is because this technique is known to be safer than the practical alternative of introducing animals. Semen is living material. There is a long list of swine pathogens that theoretically could live in it, and be transmitted through it.[27] However, as a practical matter, this doesn't seem to happen to any great extent. Maybe this is because the swine reproductive tract is not the normal route of entry for most swine pathogens. The question of whether PRRSV is an exception to this generality remains for the scientists to answer. My sense is that when all the information has been gathered and considered, PRRSV will be added to the already long list of diseases that are theoretically, but not commonly, transmitted through AI.

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