Monitoring boar studs for early detection of PRRS virus to prevent infection

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It is well documented that PRRS virus can be transmitted through semen and infect sows or gilts. The best way to prevent downstream infection is for the boar stud to remain negative to PRRS. However, if infected, early detection is critical so shipment of semen can be stopped to minimise the risk of downstream infection.

Sampling of boar studs

Most boar studs should sample blood rather than semen. It is clear the virus can be detected sooner and more readily in blood than semen in the early stages of infection in a boar.

The ideal sample is serum because the concentration of virus would be higher than in blood. Boars whose semen will be distributed downstream should be sampled at the time of collection to minimise the risk of distributing semen containing PRRS virus.

An alternative is the blood swab method. While ejaculating and without restraint, a needle prick is made in an ear vein. Using a polyester tipped swab, the blood is collected and the swab placed into a tube containing saline or in phosphate buffered saline (PBS).

Most of the universities would need a volume of 0.5ml to be able to do an individual PCR and repeat the test (in the event of a positive or suspect result).

Table 1. The number of boars that need to be tested to be 95% confident of detecting a positive boar given 5% prevalence rate.

<table>
<thead>
<tr>
<th>No. boars collected</th>
<th>No. to sample by blood/serum (pools of 5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;=30</td>
<td>Test all</td>
</tr>
<tr>
<td>31–35</td>
<td>30</td>
</tr>
<tr>
<td>36–50</td>
<td>35</td>
</tr>
<tr>
<td>51–70</td>
<td>40</td>
</tr>
<tr>
<td>71–100</td>
<td>45</td>
</tr>
<tr>
<td>101–175</td>
<td>50</td>
</tr>
<tr>
<td>176 or more</td>
<td>55</td>
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</table>

Some sensitivity is lost with this technique due to the dilution effect in the saline or PBS and also the dilution effect caused by the red blood cells.

Pooling of samples can be done in pools of three or five, understanding that some sensitivity is lost and the chances of missing a positive are greater. This is particularly true during early infection, when the odds are higher that only one positive would exist within the pool.

The blood swab method can be done on the same boars at each collection. In the field, the finch rate (% of boars responding negatively to the needle prick) has matched the research setting, at around 10%

Due to the short time frame needed to do the needle prick, the blood swab method is easier for staff when compared with drawing blood for a serum sample on an unrestrained animal.

Testing of boar studs

The simple answer is that every boar should be blood sampled at every collection and tested individually for all strains of PRRS by PCR. Of course, this is not economically feasible due to the high cost of PCR testing. Most boar studs can afford to do statistical sampling as a means of risk management downstream. For example:

- All boars whose semen will be distributed to nucleus or multiplication herds should be sampled each day they are collected.
- A statistical sample of the population collected each day should be sampled, as determined by the risk downstream farms or the stud are willing to take. The study I work with do either a 95/10 or 95/5 sample on each collection day. For a large stud the numbers approach 30 or 60 per day, respectively.
- Any boar who is off feed, feverish, or showing any clinical signs should be sampled immediately. Although clinical signs or fever by themselves are poor indicators of PRRS PCR status, a boar who has been infected with PRRS virus would be more likely to be showing clinical signs or fewer than a boar who has not been infected with PRRS virus.

Table 1 shows the number of boars that need to be tested to be 95% confident of detecting a positive boar given 5% prevalence rate.

Semens should always be withheld until negative results are obtained. There is little value in testing a stud if semen could have already been used when results are obtained.

If it is impossible to do this, it may be more appropriate to test weekly using ELISA and PCR tests, understanding that downstream infection is much more likely to have occurred when positives are found.

Economics of PCR testing

I have developed an economic impact analysis to compare testing costs related to the costs of a 200 boar stud breaking with PRRS and the boar stud supplying semen to 35,000 sows. The costs to downstream production are based on a recent publication which quantified the costs of a farm breaking with PRRS.

A summary of the costs are as follows:

- $74.16 per litter loss with PRRS break.
- $6.01 nursery loss per pig.
- $7.67 finishing loss per pig.

Based on these numbers, a 2500 sow unit would have an estimated total loss of $321,726 loss in four months. The following assumptions were made to determine this calculation:

- 17 weeks x 106 farrowings per week x $74.16 loss per farrowing.
- 17 weeks x 106 x 7.63 pigs/litter (extrapolated from same paper) x $13.68 combined nursery and finisher loss per pig.

The losses for the boar stud were estimated to be $812 per boar. I estimated this number based on multiple stud breaks that I have been involved with. An assumption was made that boars could be replaced at cost of production, which is how most boars are sold today.

An assumption was also made that 25% of downstream farms would be infected if the boar stud was only doing monthly monitoring rather than daily PCR monitoring and that stud closure would happen when clinical signs were significant enough to warrant diagnostics.

Continued on page 25
Costs of PCR testing include the following:
- $22 per PCR test.
- Blood swabs or serum tested in pools of three.
- 42 boars tested per day on average using a 95/5 statistical sampling chart based on the number of boars collected each day.
- Three collection days per week.
- Consumables (swabs, needles, microcentrifuge tubes) of $63/week.
- Extra labour of 10 hours at $12 per hour.
- Estimate of 200 miles each way to diagnostic laboratory at $.50 per mile.
- Total cost of $1707 per week.
- Total cost of $88,764 per year.

If we use the estimated total loss numbers ($1,288,441) and divide by the total cost of testing per year ($88,764), we can estimate that the stud would have to operate for 14.5 years without a PRRS break to justify not PCR testing a statistical sample on a daily basis.

Realistic expectations

Unless testing 100% of the boars every day, which in most cases is economically prohibitive, there needs to be a realistic expectation of the result.

In addition, we do not know if one animal gets infected or if more than one becomes infected at the initial disease introduction.

So, if more than one animal gets infected when the virus initially enters the farm, you will obtain different results. Table 2 shows the percentage of boars tested to be 90% confidence of finding the stud positive within two weeks after virus infected a single boar.

**Techniques for testing**
- **Semen:**
  Semen sampling should be done on raw semen only. Because of the poor sensitivity, semen samples should not be pooled for testing. They should be tested individually.
- **Blood swab:**
  Blood samples have only a slightly reduced sensitivity from serum testing. This is due to the fact that the sample needs to be diluted in saline or in phosphate buffered saline (PBS). For most of the diagnostic laboratories, about 0.5ml of sample is needed to run the necessary PCR tests to confirm an initial positive sample.

Table 2. The probability of finding virus in the stud within the first week after a single animal was infected.

<table>
<thead>
<tr>
<th>Confidence level (%)</th>
<th>Serum or blood swab (individual)</th>
<th>Serum or blood swab (pooled)</th>
<th>Semen (individual)</th>
</tr>
</thead>
<tbody>
<tr>
<td>90</td>
<td>37%</td>
<td>44%</td>
<td>74%</td>
</tr>
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</table>

As a result, we normally put the blood swab into 0.6ml of saline or PBS. It is extremely important that the swab is 100% saturated with blood.

Otherwise, the swab will soak up the saline and you will be short on sample and have potentially diluted out virus to the point a positive sample is missed. Use a swab that holds a lot of blood. The standard swab that should be used is a Quickvue inline strep A swab. It is important to use this swab because it holds about 2.5 times the amount of blood as a standard Rayon or Dacron swab.

- **Semen:**
  Serum is the ideal sample to use. PRRS virus is in much higher levels in the serum than in semen and at least 1-2 days sooner. Serum samples can be taken from the ear vein (auricular vein), leg vein (saphenous vein), or tail vein. Sampling is much easier if done while the boar is ejaculating. Another option is to retrain the boar and bleed from the neck.

**PRRS ELISA testing**

Even with extensive PCR testing, there is the chance that the PCR test will not detect the strain of virus that has infected a herd. This has happened, and in one case a new, undetectable (by PCR) strain was introduced into a boar stud. For this reason, it is still important to do PRRS ELISA testing.

The ELISA test detects antibodies about two weeks after an animal has been exposed to PRRS virus, but it is extremely sensitive to all PRRS virus strains. It is recommended that this is done on a weekly basis. Most studs do about 30 boars throughout a monthly period.

**Summary**

Boars are positive in serum/blood before semen by PCR and in much higher quantity in the early stages of infection, thus blood sampling is the preferred method for monitoring of negative studs. Monitoring should occur daily. Choosing how many boars to test and which boars to test should be determined on an individual system basis. This is dependent on the estimated risk of being infected and the potential negative costs of downstream infection. Semen should always be withheld prior to when PCR negative results are obtained.

References are available from the author on request.