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PRRS Ctrl 2.0

Plan, Check, Manage & Improve

New Strategies for PRRSv Management

Controlling PRRS has huge upsides in production economics but can be a challenging task. Follow this structured and effective PRRS control plan. Start implement a cost effective monitoring and apply customized actions to move PRRS out of your system.

This practical guidebook, combines latest field applicable research and past experiences to successfully control this disease.





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Veterinary Degree and MBA in Brazil
PhD in Veterinary Population Medicine in the US

Worked for 8 years in the industry on health services, product validation, supply chain, and technical services. Since 2015 he is an Associate Professor and Director of Graduate Education at Iowa State University.

Leads a team of multiple graduate students working towards improving health & productivity of swine populations under field conditions. His group has over 300 publications/participations on meetings over the last 5 years, having worked closely with veterinarians and pig producers in Canada, USA, Mexico, China, Spain and Brazil.



Dr. Marius Kunze
Veterinarian

Before joining Boehringer Ingelheim in 2014, I started my career as a pathologist focusing on swine diseases. After working as a technical service veterinarian, my responsibilities have shifted, since 2019 I support globally our PRRS and PPV brand.

A great benefit of my profession is to help producers improving their performance with innovative and flexible solutions in disease control. Collaborating with Global swine experts, and share the outcome of their field applicable research with veterinarians and producers is an important and fun part of my job.

Marius and I hope you enjoy this book as much as we do. We've made effort to make it simple, straightforward, and practical with field-applicable information. Please reach out to us should you want to further discuss any topic covered here.

Happy reading! All the best,

Introduction to the document

PRRSv is an ever-evolving virus affecting the global swine industry. There have been several exciting new developments to help veterinarians and producers implementing strategies to manage PRRSv infection.

This document provides a summary of monitoring and surveillance systems (MOSS) available using ongoing monitoring of production & clinical data, as well as using diagnostics. There are new tools and strategies in both fronts, which are presented here. The information from these MOSS tools allows early detection of outbreaks and classifying herds according to PRRSv activity. These measurements allow for a more accurate evaluation of progress towards virus control and elimination.

The first part of the document dives into the new MOSS tools at the pig level (i.e., using serum or oral swabs) or the population level (i.e., using processing fluids, tongue tips fluids, oral fluids). We present the tools, their application, and a guide on how to interpret results related to PRRSv activity, and how to associate that with PRRSv management strategies (control or elimination).

The second part provides a guide on how to use the MOSS results to classify herds according to PRRSv exposure and active circulation. This facilitates communication within and between production systems and veterinary clinics, allows veterinarians and producers to measure the effectiveness of different interventions to prevent, detect, or manage PRRSv infection; and also allows academic institutions to conduct standardized epidemiological studies and economic assessment at the system, region, or national levels on PRRSv impact on productivity.

The third chapter provides insights on key factors to manage PRRSv infection in affected herds, sharing results from field-based studies on factors associated to shorten the time to produce negative piglets and to minimize the production losses incurred from PRRSv infection.

Altogether, the document provides field-based data on aspects and strategies associated with response & recovery of PRRSv infection, helping veterinarians and producers keep improving efforts to improve animal health & productivity.

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Classifying PRRSv status: establishing exposure & shedding at the herd level

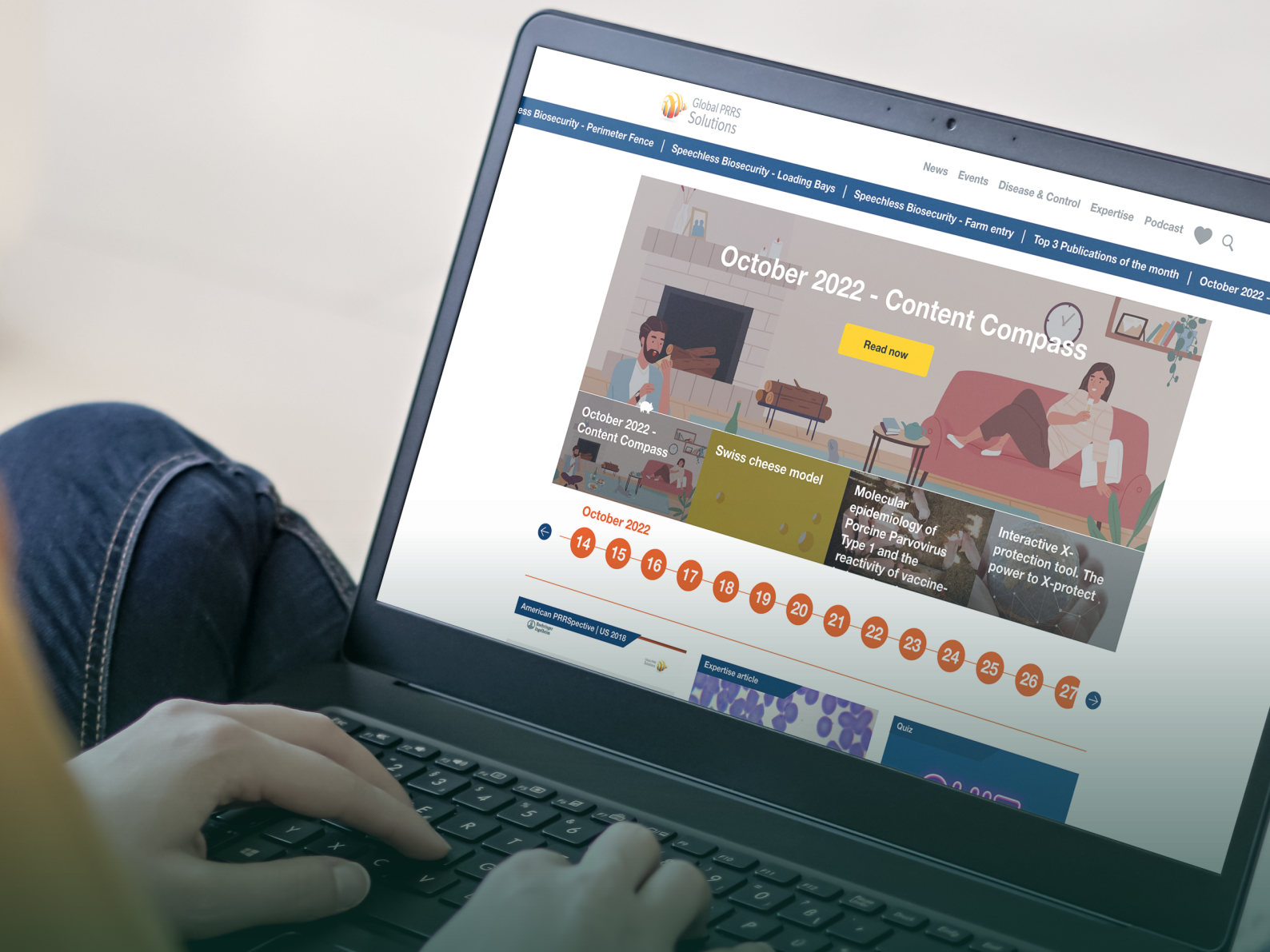
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1.0 Monitoring and Surveillance Systems available



Why monitoring herds for PRRSv?

- PRRSv status is a great predictor of productivity. The level of PRRSv shedding correlates very well with parameters such as feed intake from sows, number of abortions, farrowing rate, neonatal losses, pre-weaning mortality, and number of pigs weaned (*Figure 1*). In the grow-finish area, it correlates with feed intake, average daily gain, mortality, and antimicrobial use, worsening all parameters listed. Therefore, closely tracking PRRSv status of breeding herds allows systems to fine tune their herd management, pig flow, and immunization strategies to improve (or maintain) status over time, maximizing the whole herd health and productivity (*Figure 1*).
- For PRRSv-negative herds, the earlier outbreaks are detected, the earlier veterinarians can recommend strategies. Also, for production system having multiple herds, early detection of outbreaks allows preventing further virus spread between herds by shared personnel and equipment, and pig movement. For endemically infected herds, it is still crucial to monitor for PRRSv activity, understanding if the predominantly circulating viruses are MLV-like or wild type. If wild-type, is there diversity over time, or is it a similar family of viruses circulating?



Moura *et al.*, 2022

Is prevalence of PRRSv low, medium, or high, and is it increasing, decreasing, or stable over time? Understanding these questions allows defining the need to improve biosecurity (prevent unrelated virus introductions), or bio-management strategies including gilt flow and herd immunization, leading to decreasing prevalence and activity of wild-type viruses. Also, the level of activity is associated with productivity. The higher the prevalence of wild-type, the lower productivity is expected from the affected herds (Moura *et al.*, 2022).

- Some systems need to commingle flows of different sources in the same grow-finish barn. Commingling flows of similar PRRSv status allows standardizing preventive or therapeutic solutions, and minimizes losses. Mixing acutely infected flows with negative or low-prevalence flows leads to more virus circulation and impact on the herd's health and productivity.
- Tracking PRRSv activity over time also allows veterinarians to measure effectiveness of biosecurity, bio-management, and bio-containment measures in place. It, therefore, provides a roadmap to evaluate progress of whole-system, regional, or national control measures.
- Moreover, tracking PRRSv status over time allows measuring disease incidence, which are great indicators of overall biosecurity.

Figure 1. Control charts of the rate of regular returns to estrus (A) and the pregnancy rate (B) before and after implementation of a whole herd vaccination program against PRRSv in a 2300 head sow farm in Serbia with a history of low farrowing rates.

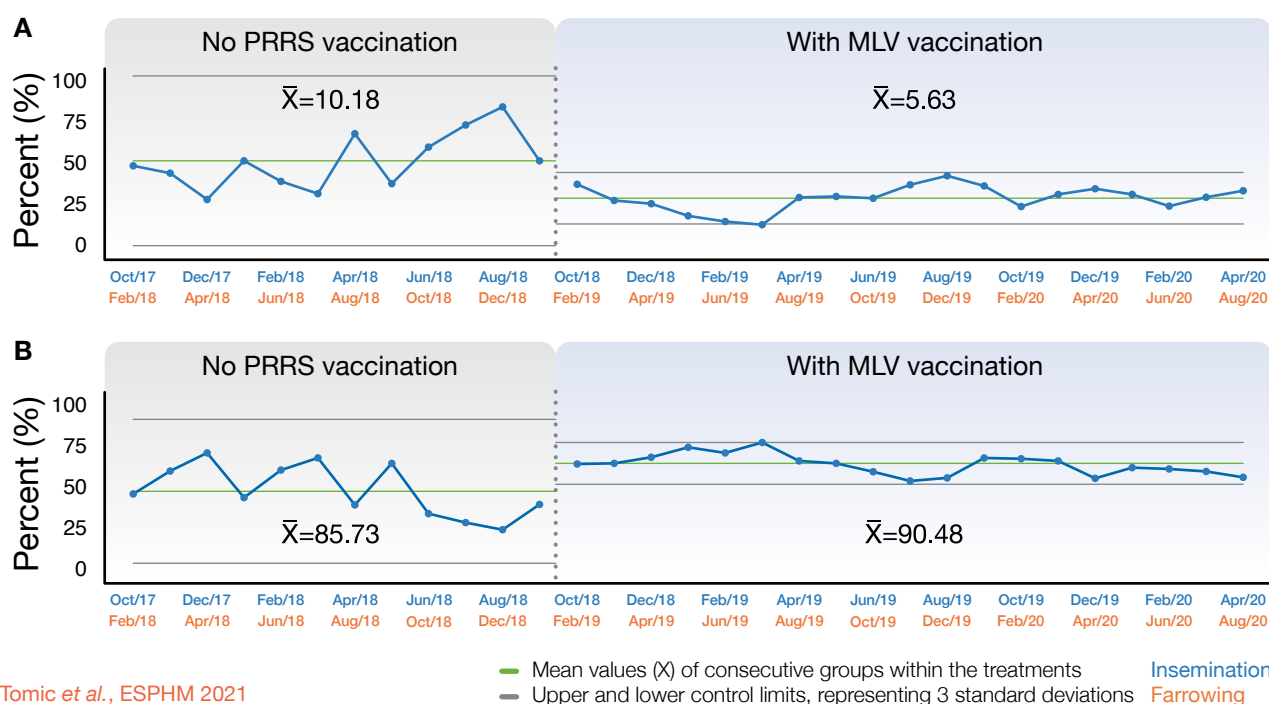
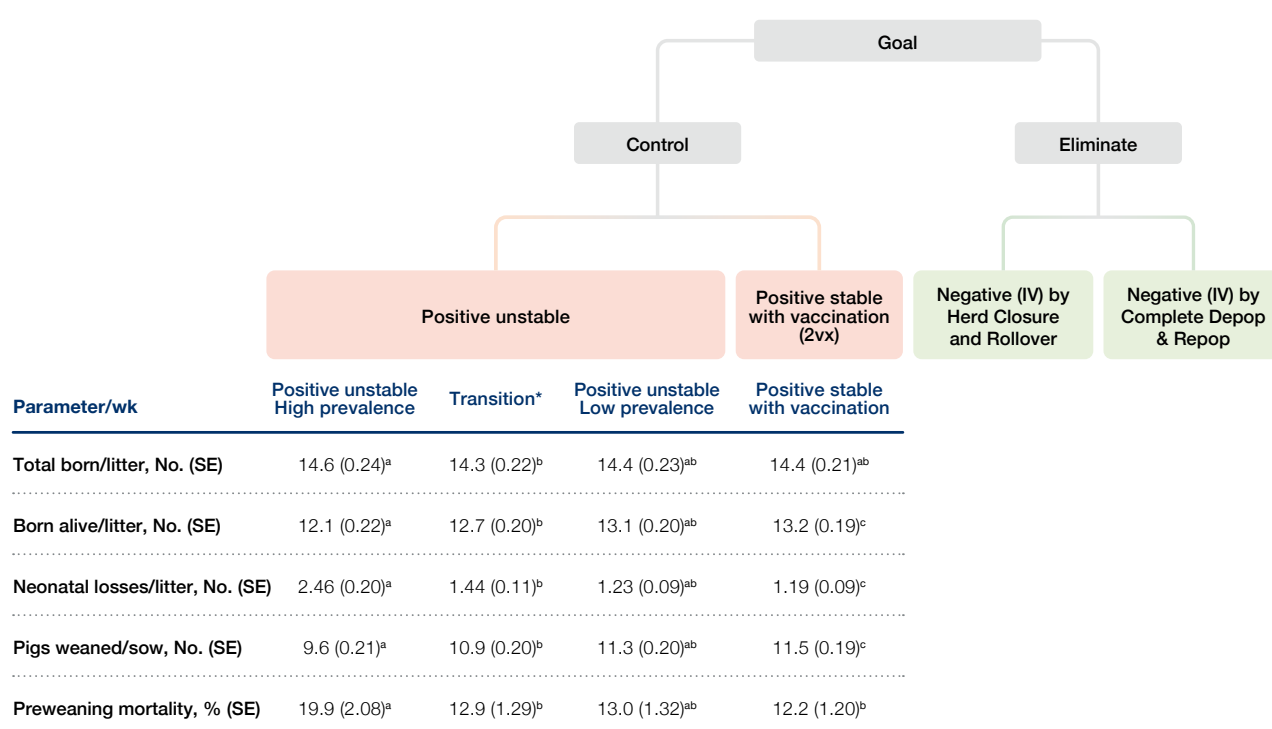


Figure 2. PRRS status and reproductive performance.

Farm data over 4 years were collected and matched with PRRS classification. Productivity changes were characterized as herds transitioned through categories. Productivity improved as farm status improved.



* This period begins on the 11th week of a herd being classified as 1A status post-PRRSv outbreak and ends when the herd was promoted to 1B status.

^{a,b,c} Different superscripts on compared statuses for each productivity parameter indicate statistical differences ($\alpha = .05$).

* SE: Least-squares means (SE) of productivity parameters for each AASV 2.0 PRRSv status classification.



Holtkamp *et al.*, 2013



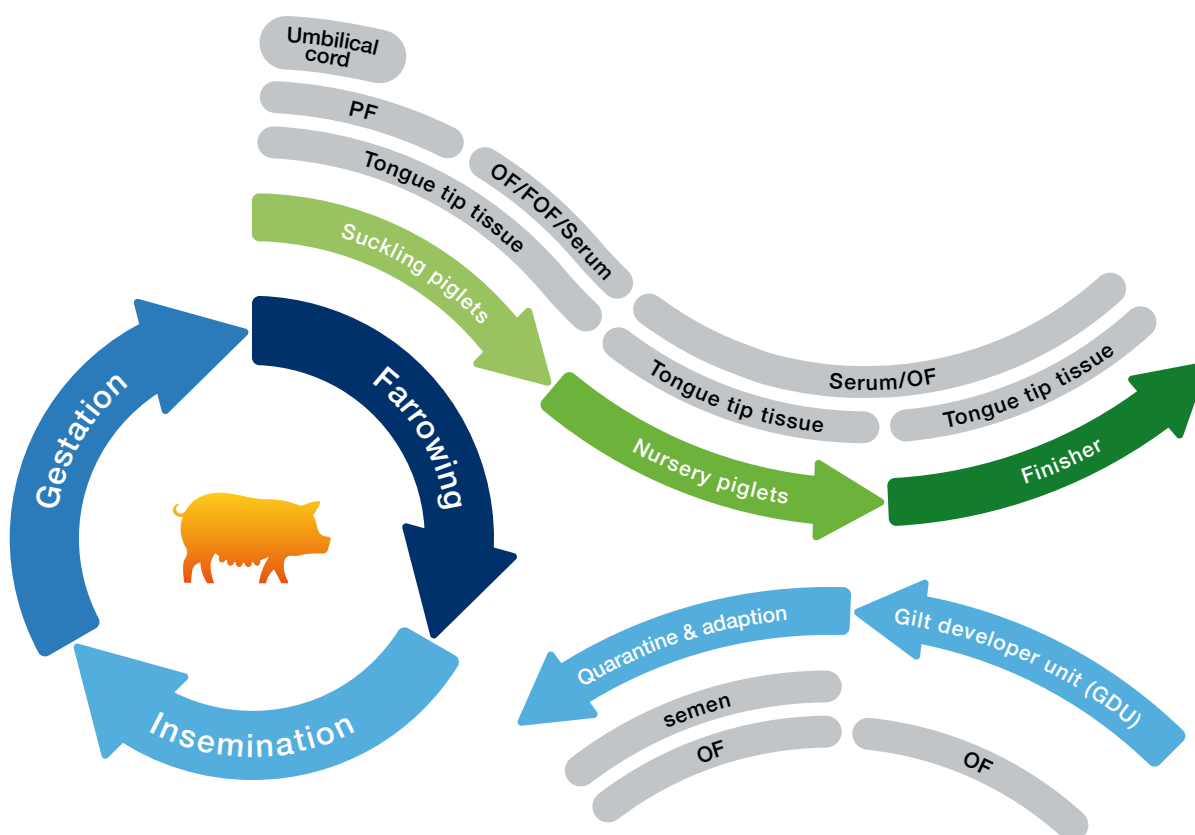
Osemeke *et al.*, 2021

1.1 Monitoring and surveillance systems for PRRSv

In the early 2000's, testing blood samples was the only available method to monitor PRRSv in pig populations. Since then, several other population methods have been developed. **Processing fluids (PF)** is an aggregated sample type allowing efficiently monitoring hundreds of <5 days old piglets. **Family oral fluids (FOF)** can be applied to weaning age pigs, reflecting the PRRSv activity in weaning litters. **Oral fluids (OF)** can be collected from pigs of any age starting at weaning, and including sows and boars. More recently, **Tongue tips fluids (TTF)** have been reported as a targeted sample collected from dead animals of any age group (Figure 3).

Considering that diagnostic monitoring is not done on a daily basis in most pig herds, production data is a great complement to diagnostic monitoring, allowing people to screen herds for signals (i.e., spike in abortions, or drop in the volume of pigs weaned) commonly associated with disease outbreaks (Figure 4).

Figure 3. Samples used for PRRSv detection.



1.1.1 Clinical and production data: Early outbreak identification

Age groups: Production data allows monitoring disease activity in multiple production stages

Clinical data: Number of sows off feed.

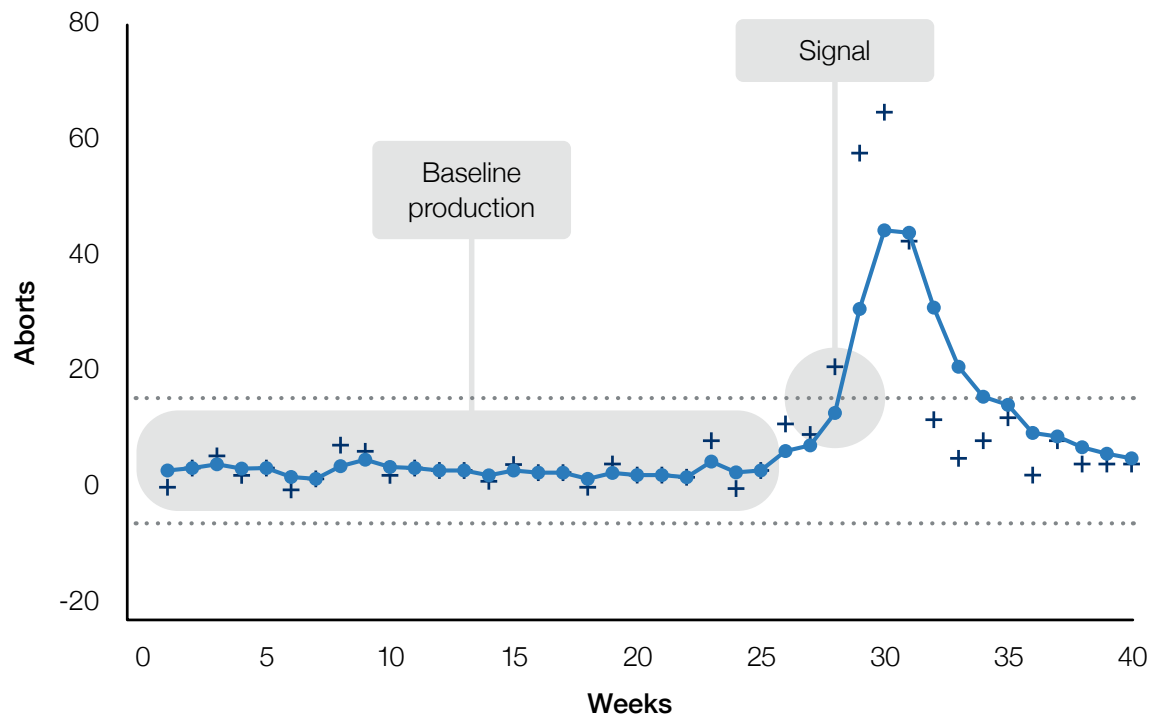
Production data: Number of abortions, prenatal losses, pre-weaning mortality, sow mortality.

Pro Easy to collect, daily updates, complement routine diagnostic monitoring, cheap and practical tool for early detection.

Caution Need algorithms (define signals), require “digital” data, change in productivity are not disease-specific, required follow-up diagnostics to define the cause(s) of variation.

Action items Put the herd on hold until diagnostics work-up reveals the causes of variation.

Figure 4. Number of abortions in infected farm with 1000 sows.



1.1.1 Clinical and production data: Early outbreak identification

Most farms report changes in productivity following PRRSv outbreaks.

Most sensitive indicators:

Number of sows off-feed in the gestation line (2+ weeks post service) and number of abortions.

Also impacted (with some delay):

Pre-weaning mortality and Neonatal losses (mummies, stillbirths).

Note the variability on changes in productivity between herds. Factors such as herd immunity at the time of the outbreak and virulence of the PRRSv infecting the herd play a big role in the changes in productivity following an outbreak. This is why it is advised to monitor multiple parameters over time.



Figure 5. Pre-weaning mortality over time in 10 breed-to-wean herds.

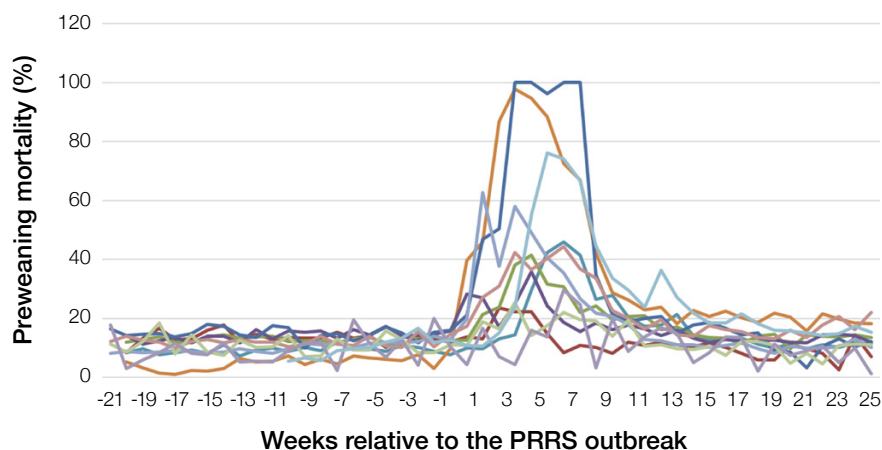
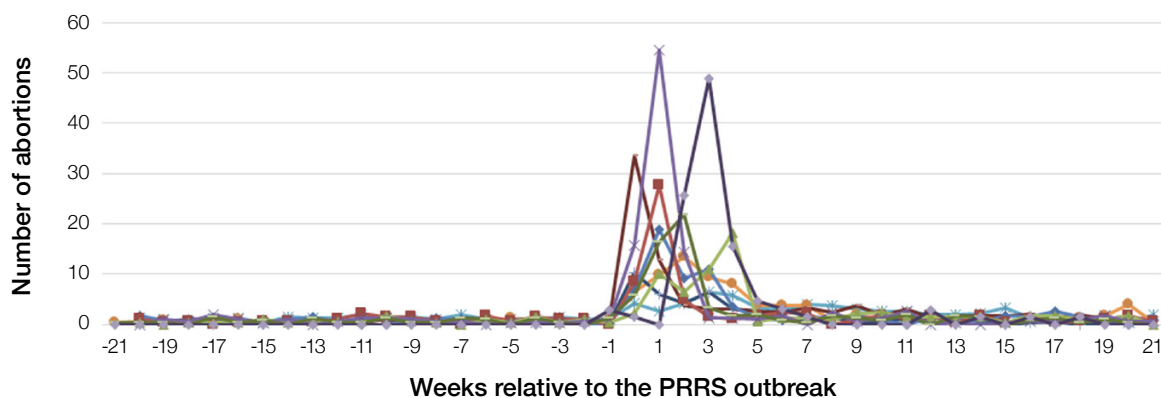


Figure 6. Number of abortions per 1.000 sows over time in 10 breed-to-wean herds.



1.1.2 Serum samples

Gold-standard to define viremia

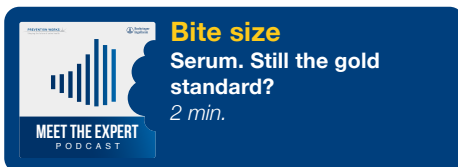
Age groups: Pigs of all ages.

Pro Most people are familiar, reliable results, good for serology (ELISA, IFA), for simple molecular tests (PCR) and for advanced molecular (sequencing) test. Also great to establish prevalence at the pen, room, or whole-barn levels.

Caution Requires training people, causes stress (sometimes mortality) in pigs, spread pathogens when collecting multiple pigs, for great herd sensitivity at low prevalence this method requires a massive sample size.

Action items

- **Positive results on ELISA** = evidence of past or recent PRRSv exposure.
- **Positive results on PCR** = evidence of virus ongoing circulation.



1.1.2 Serum samples

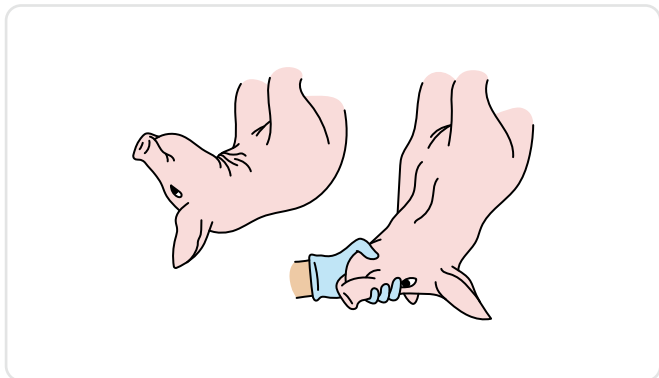
Sampling technique



Adapted from:
www.securepork.org

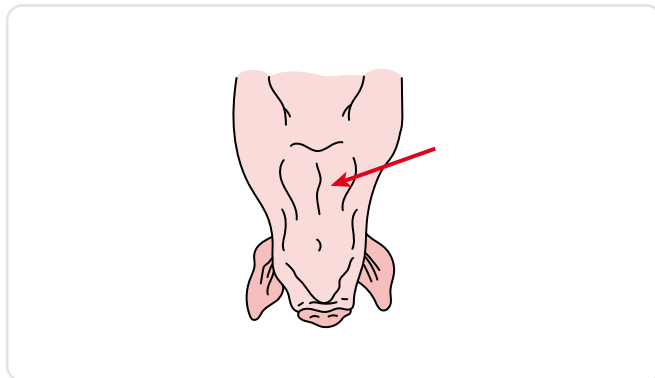
STEP 1

Person collecting blood, use non-bleeding hand to gently extend neck, being aware to not restrict breathing.



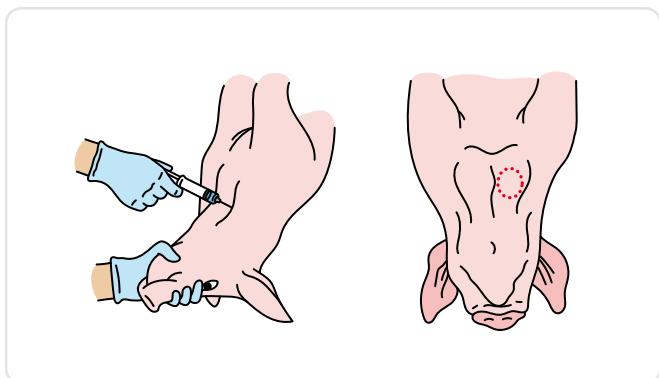
STEP 2

Collect blood from jugular groove with a blind stick, starting on the pig's right side preferably.



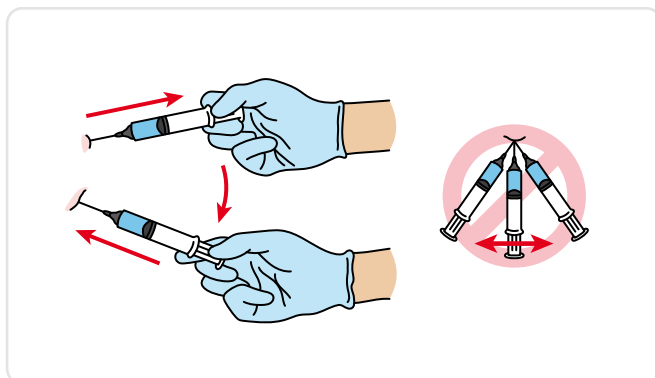
STEP 3

Ensure needle is perpendicular to the skin. The deepest part of the jugular groove is the entry point.



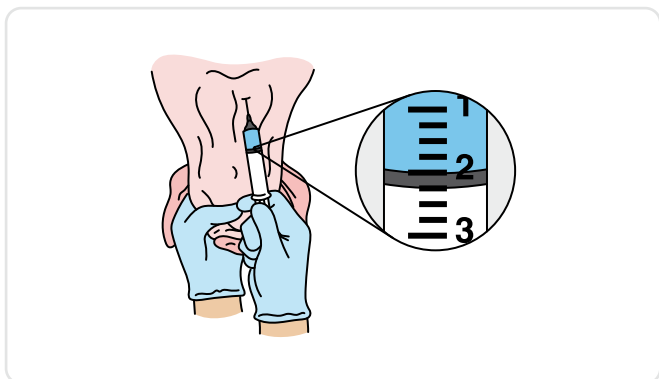
STEP 4

Adjust needle angle and depth until blood flows. To reposition, pull back, adjust angle, increase or decrease depth.



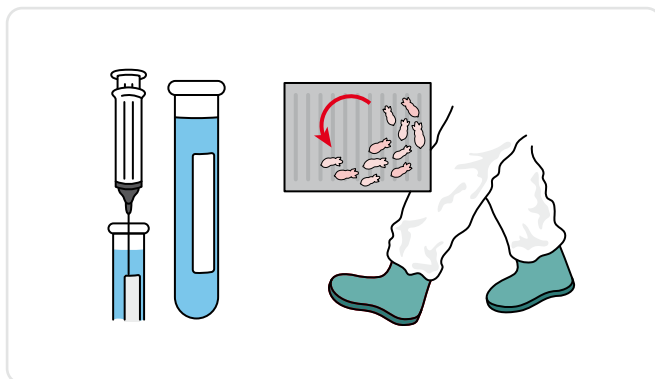
STEP 5

Once blood is flowing, collect a minimum of 2 ml for most diagnostic test. For future reference, note the position and depth before removing the needle.



STEP 6

Transfer blood from syringe to blood tube. Check animals to confirm normal activity in pen when bleeding is complete. Promptly chill* samples for shipment.



*Only freeze serum not whole blood.

When a litter is PRRSv-positive, how many piglets (within the litter) are usually positive?

There are litters with several positive pigs. However, especially when prevalence is low, the most common is to find just a few pigs viremic in the litter. The rest will likely be negative on serum PCR.

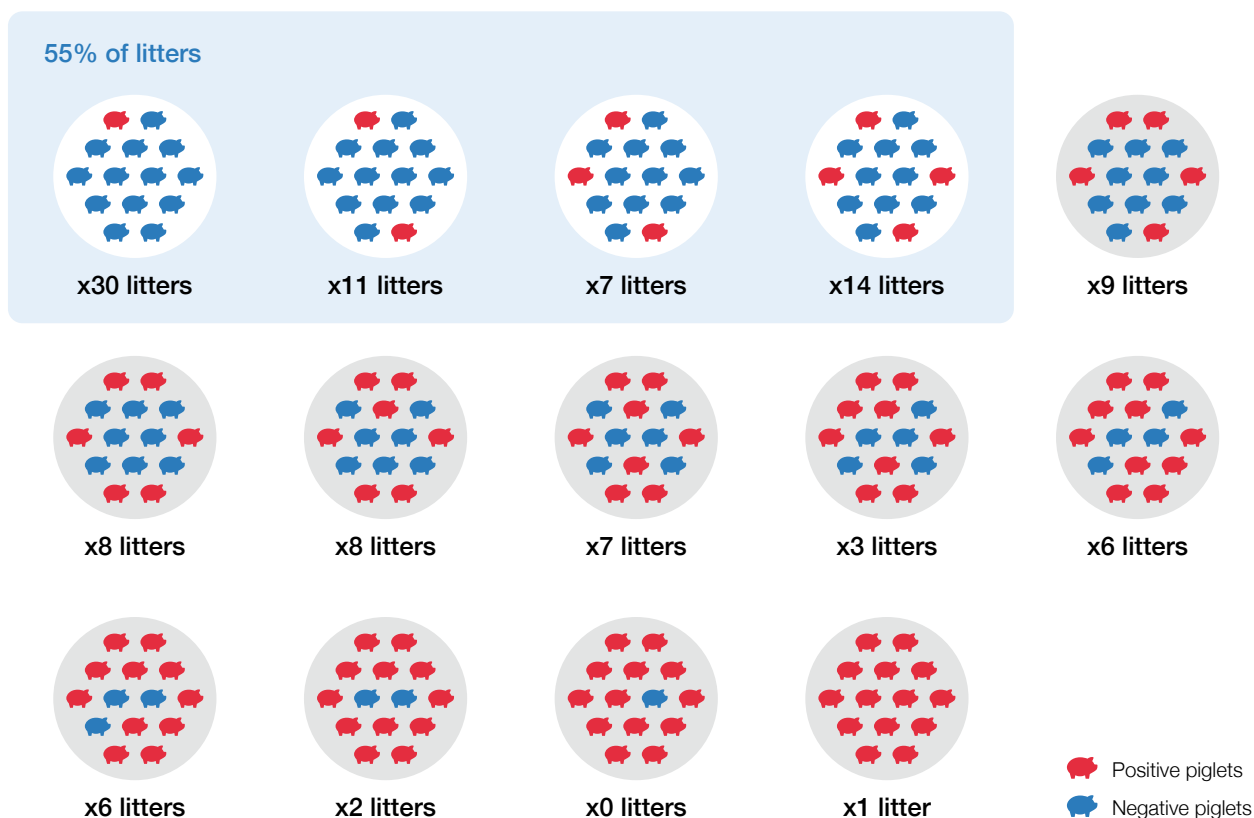
In practical terms, this means that **individual pig sampling is not efficient** in low prevalence scenario, as it requires a large sample size (*Table 2*).

Alternatively, **family oral fluids (FOF) can efficiently detect virus** using far less number of PCR tests.

Example: 90 serum samples or 10 FOF, per air space, is needed to achieve 95% confidence to detect at least 1 sample positive when prevalence is 3% or higher.

Figure 7. Number of PRRSv-viremic piglets in breeding herd.

A cross-sectional study was performed in **12 breed-to-wean sow farms** in which serum samples ($n = 4510$) were collected from all piglets in selected litters ($n = 422$) in **23 farrowing rooms** and tested individually for PRRSv RNA. In total, **112 litters were tested positive**. This image below shows how PRRSv was distributed in the these positive litters.



When a litter is PRRSv-positive, how many piglets (within the litter) are usually positive?

In general, serum sampling is the gold standard: most accurate, and better quality sample for several basic and advanced diagnostic tests.

However, in low-prevalence scenarios it requires a large (120+) sample size which can be time and cost-prohibitive.



fieldepi.org

Table 1. Comparison of attributes and key features from serum, processing fluids, and family oral fluids for PRRSv monitoring in breeding herds.

Serum	Tongue Tips Fluids (TTF)	Processing Fluids (PF)	Family Oral Fluids (FOF)
Classic approach: people know how to do it	Risk-based monitoring of pigs of different age groups, bags ~ 20–100 tongue tips/bag	Simple and effective way to screen populations	Require more work than PF, less than serum
Great to determine prevalence	<ul style="list-style-type: none"> Gauging vertical transmission: stillbirth tongue tip samples Assessing PRRSv activity across rooms between processing and weaning 	Great herd sensitivity: <ul style="list-style-type: none"> 1 PF/whole week At near-zero prevalence: 1 PF per ~ 30 litters 	10 FOF = 95% confidence to detect PRRSv at low prevalence. Pool samples 1:5 or 1:10 when sample the whole room
Ideal for <i>advanced</i> diagnostics	Used for molecular and ELISA testing	Perhaps the most cost-effective virus circulation screening method	Can be used for detection, or to estimate prevalence
Any age	Neonates, ages outside the PF and FOF window	2–5 old piglets, mostly males	≥ 18 days old piglets (confirm due to wean (DTW) piglet PRRSv status)



Regardless of the specimen used, remember that there is a great variability of virus activity within litters, crates, and rooms. Therefore, it is important that sampling must be done repeatedly over time & geographic space. In other words, caution while extrapolating results from one sampling event to other rooms, the whole herd, or to several weeks going forward. PRRSv prevalence is very dynamic and ever changing within different air spaces in swine populations.

When a litter is PRRSv-positive, how many piglets (within the litter) are usually positive?

And how many samples do I need to collect to detect PRRSv at different prevalences (95% confidence)?

Table 2. Number of serum and FOF samples to achieve 95% confidence to detect PRRSv at different prevalence scenarios.

Prevalence (%)	# Serum samples	# FOF samples
~9	30	5
~5	60	7
~3	90	10
~2	120	15
~1	240	30
~0.5	400	40

Example: 90 serum samples or 10 FOF, per air space, is needed to achieve 95% confidence to detect at least 1 sample positive when prevalence is 3% or higher.



Note the high sample size required for serum compared to FOF for all prevalence scenarios. One of the reasons is that FOF includes biological sample from multiple animals.

1.1.3 Processing Fluids (PF)

Age group: 2-5 days old piglets.

Pro High sensitivity at whole room level; easy, practical, "the new norm" in several places, can be used for serology (ELISA) and basic molecular (PCR) testing.

Caution Not great for advanced molecular testing (e.g., NGS), PRRS status 3-5 days of life may not reflect whole-herd or due-to-wean population. The great sensitivity of PF comes from testicle tissues, blood, and lymphoid fluids. Thus, for herds not castrating (only tail docking), PF will have significantly lower sensitivity.

Action items

- **Positive results on ELISA** = evidence of PRRSv exposure.
- **Positive results on PCR** = evidence of virus circulation around birth days.

Usually PF are great for screening for PRRSv. When a few negative tests are obtained on weekly testing, a weaning -age testing is recommended to assure negative status.



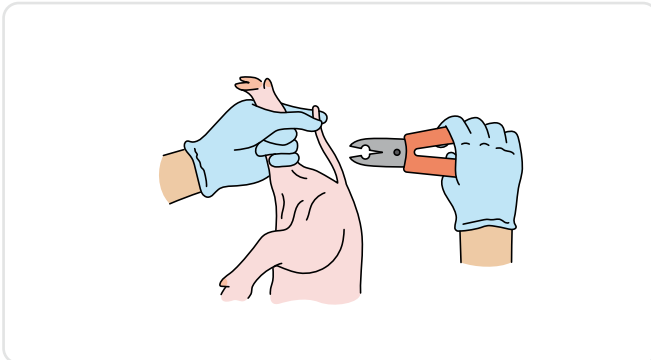
Cheesecloth with tails and testicles. Courtesy: Will Lopez.

1.1.3 Processing Fluids (PF)

Sampling technique

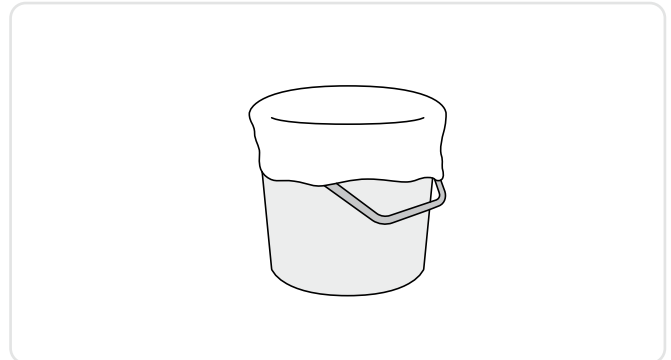
STEP 1

While processing piglets, place tails and testicles in a clean plastic bag.



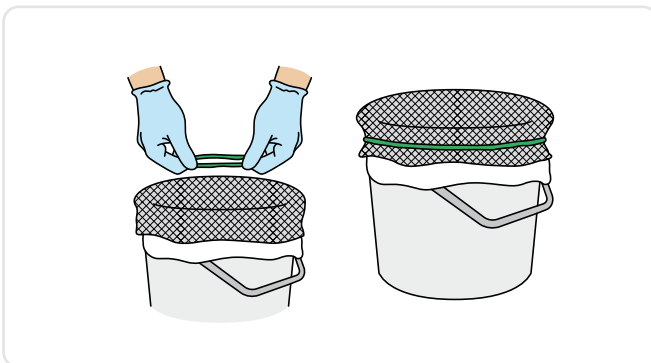
STEP 2

Line a container with a clean plastic bag.



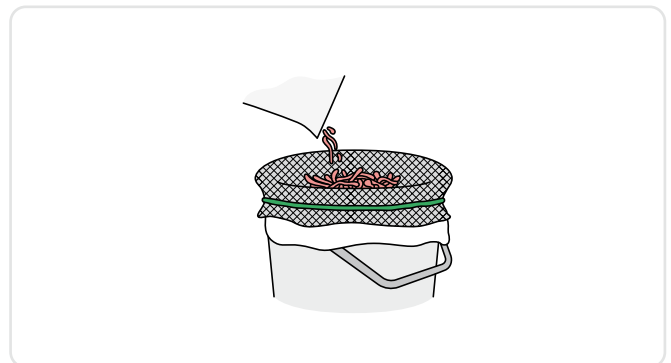
STEP 3

Place cheesecloth over opening of container. Secure cheesecloth and plastic bag with rubber band.



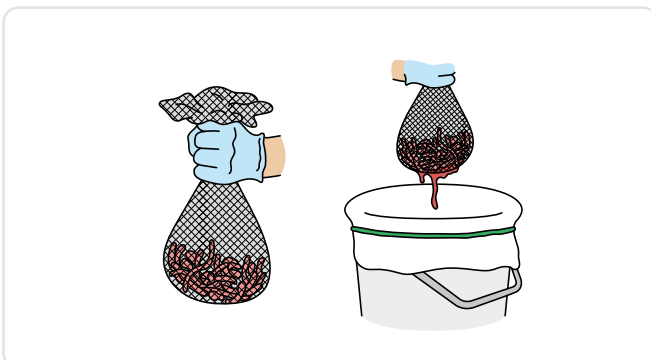
STEP 4

Pour tissues onto cheesecloth. Allow fluids to drain into the container.



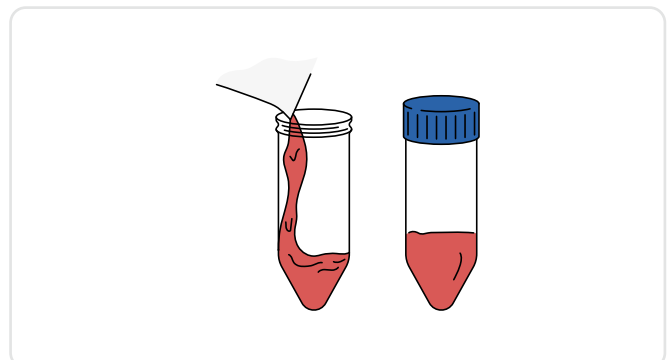
STEP 5

Gather each end of cheesecloth so tissues sit in the center, apply gentle pressure.



STEP 6

Transfer fluids from bag to conical tube. Promptly chill or freeze samples for shipment.



Adapted from:
www.securepork.org

1.1.3 Processing Fluids (PF)

The evolution of specimens

This is from the Swine Disease Reporting System, a Project that collects and aggregates diagnostic data from 5 Veterinary Diagnostic Laboratories in the US (Iowa, Minnesota, South Dakota, Kansas, Ohio).

Serum samples (~50%) and tissues (38%) were the predominant sample types until 2008, when oral fluids (OF) emerged.

2012: “Alternative” sample types started to pick up including OF and blood swab.

2017: Processing fluids (PF) emerged and quickly became the most predominant sample type (63% samples).

Serum and oral fluid (OF) are still being done, majority to:
A. Confirm status of weaning-age piglets.
B. For advanced molecular diagnostics.

OF is still the most predominant sample when considering all age groups. It is widely used in grow-finish and gilt development units for PRRSv screening.



fieldepi.org

Figure 8. Specimens submitted for PRRS virus PCR in growing pigs.

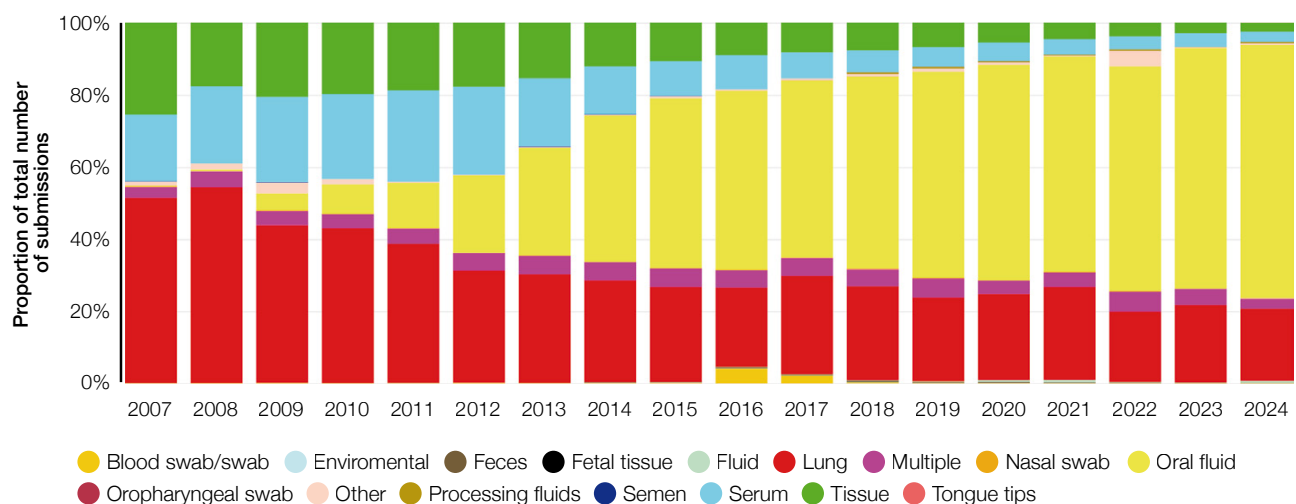
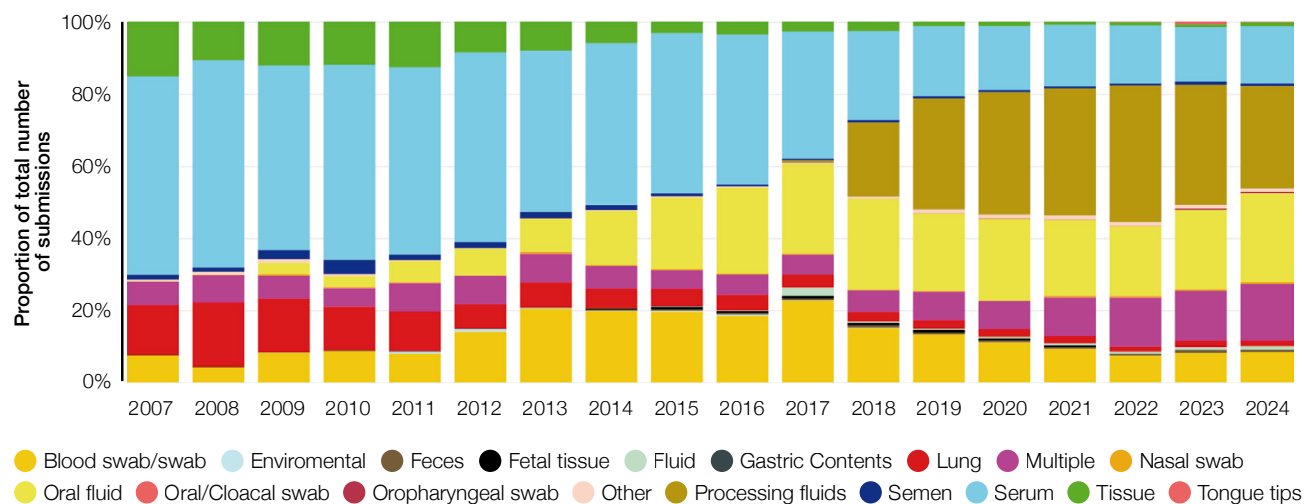


Figure 9. Specimens submitted for PRRS virus PCR in sows.



1.1.3 Processing Fluids (PF)

Amongst all specimens, PF is the most efficient to detect PRRSv RNA by PCR. This is not surprising, as this sample type consists of blood fluids, testicle tissues and fluids, and lymphoid fluids. Those are rich in cells of the macrophage lineage, which is where PRRSv replicates.

Regardless of the specimen used, remember that there is a great variability of virus activity within litters, crates, and rooms. Therefore, it is important that sampling must be done repeatedly over time & geographic space. In other words, caution while extrapolating results from one sampling event to other rooms, the whole herd, or to several weeks going forward. PRRSv prevalence is very dynamic and ever changing within different air spaces in swine populations.

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Any age	Neonates, ages outside the PF and FOF window	2–5 old piglets, mostly males	≥ 18 days old piglets (confirm due to wean (DTW) piglet PRRSv status)

1.1.3 Processing Fluids (PF)

Case report



Use of processing fluid for monitoring the status of a PRRSv outbreak in a 3000-sow herd.

Case history

A PRRS naïve 3000 sow farm located in Germany, broke with PRRS type 1 in February 2019. Clinical symptoms were most prominent in suckling piglets

with a high percentage of weak born piglets and mortality rising up to 25%.

Secondary bacterial infections were dominant in nursery pigs with increased mortality up to 10%. Late term abortions in sows reached 2%.

From April onwards, sows were vaccinated twice, four weeks apart, and piglets at three weeks of age with a PRRS MLV vaccine.

Five weeks after sow herd vaccination, every four-weeks processing fluid (4x ten litters of tails and testicles) and serum in 3 weeks old piglets were collected.



Example picture of PRRS positive weak born piglets. Courtesy: E. Marco.

1.1.3 Processing Fluids (PF)

Results

Table 3. PRRSv detection (PCR) in processing fluids.

After starting the PRRS control approach (whole herd vaccination and Biosecurity improvements) in April, virus load in processing fluids decreased in July. From September onward PRRS stability was achieved and maintained.

Month (2019)	Number of samples (PF)	Positive	Negative	Max viral load, log GE/ml
April	4	4	0	5,21
May	6	6	0	6,71
June	7	1	6	–
July	15	6	9	4,97
August	19	2	17	4,65
September	14	0	14	0
October	13	2	11	–

Table 4. PRRSv detection (PCR) in serum.

Serum samples in 3 week old pigs (prior to vaccination) showed matching results compared to processing fluid samples in same animals.

Date	Number of samples (pools)	Positive pools	Negative pools	Max viral load, log GE/ml
5.2.2019	30 (6)	6	0	5,98
5.23.2019	30 (6)	3	3	4,24
6.13.2019	30 (6)	2	4	4,75
7.24.2019	30 (6)	0	6	–
9.11.2019	30 (6)	5	1	4,92
9.26.2019	30 (6)	0	6	–
10.17.2019	30 (6)	0	6	–
10.24.2019	20 (4)	0	4	–

Conclusions

- Gilt introduction was the likely source of infection in a previously naive 3000 sow farm.
- Most prominent clinical symptoms were weak born piglets, increased mortality and secondary bacterial infections.
- After the outbreak, whole herd vaccination was implemented (Sow and piglet vaccination). According to the outcome of a personalized biosecurity assessment, actions were adapted and implemented.
- Monitoring the PRRSv prevalence after the outbreak with processing fluids, helped to identify further biosecurity breaches.
- 35 weeks after the outbreak, time to baseline production was achieved.

1.1.4 Family oral fluids (FOF)

Age group: prior to weaning.

Pro High sensitivity at whole room level; easy, practical, can be used for serology (ELISA) and basic molecular (PCR) testing; require much less sample size than serum to achieve the same confidence.

Caution As FOF is used to confirm negative status of weaning-age litters, it usually yields high Cts (>30), so usually are not great for advanced molecular testing (e.g., next generation sequencing). Gives result at litter level (not individual animals). Not always in agreement with PF-monitoring.

Action items

- **Positive results on ELISA** = evidence of PRRSv exposure.
- **Positive results on PCR** = evidence of virus circulation around weaning.



1.1.4 Family oral fluids (FOF)

Sampling technique



Success rate goes up when FOF is collected as early in the morning as possible (active sows getting up to eat).

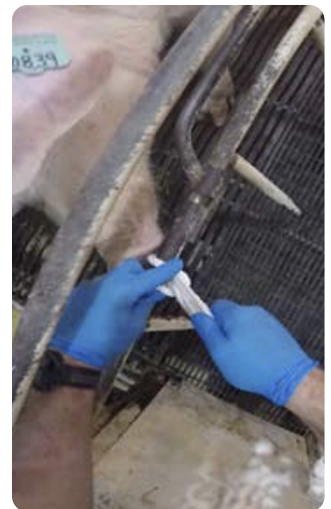
STEP 1

Untangle a bleach-free cotton rope, so that you have three smaller pieces readily accessible to the piglets.



STEP 2

Tie the rope to the front of the crate, on the opposite side of the sow's water. Hang it so it is ~ 1–2 inches / 2–3 cm from the floor.



STEP 3

Secure the rope using a zip tie, making sure the sow won't be able to untie it.



STEP 4

Leave the rope there for ~ 30 minutes, then squeeze the contents of the rope into a clean zip-lock bag (you can do this while the rope is still tied to the metal part of the crate) and transfer it into a tube. Cut the zip tie, remove the rope, and discard it. Since we are pooling the samples by room, no need to change gloves inside the same farrowing room, but make sure you change gloves between air spaces.



Pictures and instructions: Dr. Marcelo Almeida, ISU FieldEpi team.

1.1.4 Family oral fluids (FOF): High herd sensitivity to detect PRRSv in piglets at weaning

This data is from a study with 72 matching sets of family oral fluids (FOF) and sera from all piglets in each litter.

FOF and each serum was individually tested for PRRSv RNA by rRT-PCR. Every time family oral fluids was negative, there were no viremic piglets in the litter.

This means the Specificity of FOF was 100%. In other words, no false-positive for FOF, i.e., a PCR-positive FOF means there was a viremic piglet in the litter.

As the number of viremic piglets increased in the litter, the higher was the probability of detecting PRRSv in FOF.

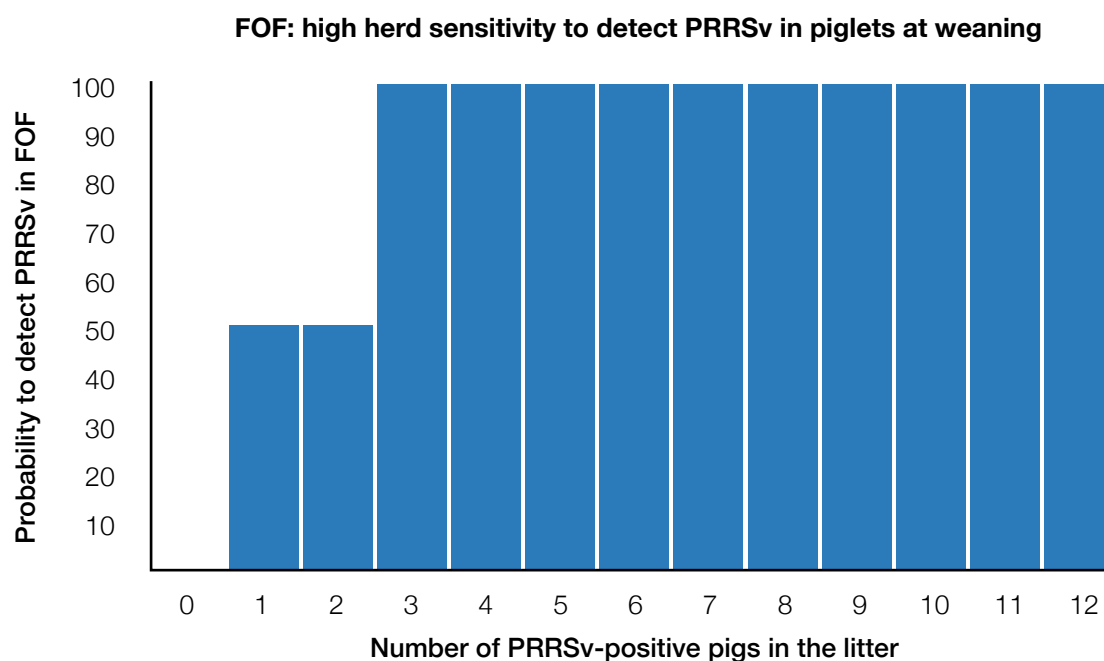


Almeida *et al.*, 2019



50% chance when 1–2 viremic piglets.
100% chance when 3+ viremic piglets in the litter.

Figure 10. Probability for PRRSv detection and number of positive piglets per litter.



1.1.4 Family oral fluids (FOF): High herd sensitivity to detect PRRSv in piglets at weaning

FOF is not as practical nor efficient as PF to detect PRRSv. However, it is significantly easier and more efficient than individual pig testing using serum or blood swabs (see the table at the bottom of this page with sample sizes for PRRSv monitoring in weaning-age pigs).

For example, to detect PRRSv at 1% prevalence, there is the need of 240 serum samples, or 30 FOF. If submitting this 30 FOFs in pools of 1:10, that means 3 PCR tests.



Regardless of the specimen used, remember that there is a great variability of virus activity within litters, crates, and rooms. Therefore, it is important that sampling must be done repeatedly over time & geographic space. In other words, caution while extrapolating results from one sampling event to other rooms, the whole herd, or to several weeks going forward. PRRSv prevalence is very dynamic and ever changing within different air spaces in swine populations.

Table 5. Comparison of attributes and key features from serum, processing fluids, and family oral fluids for PRRSv monitoring in breeding herds.

Serum	Processing Fluids (PF)	Family Oral Fluids (FOF)
Classic approach: people know how to do it	Simple and effective way to screen populations	Require more work than PF, less than serum
Great to determine prevalence	Great herd sensitivity: <ul style="list-style-type: none"> 1 PF/whole week until testing negative by PCR. When near-zero prevalence, use 1 PF sample per ~ 30-35 litters 	10 FOF = 95% confidence to detect PRRSv at low prevalence. Ok to pool 1:5 or 1:10 when sample the whole room
Ideal for <i>advanced</i> diagnostics	Perhaps the most cost-effective virus circulation screening method	Can be used for detection, or to estimate prevalence
Any age	2-5 old piglets, mostly males	≥ 18 days old piglets (confirm due to wean (DTW) piglet PRRSv status)

Number of serum and family oral fluid (FOF) samples to achieve 95% confidence to detect PRRSv at different prevalence scenarios.

Prevalence (%)	No. Serum samples	No. FOF samples
~9	30	5
~5	60	7
~3	90	10
~2	120	15
~1	240	30
~0.5	400	40

*** Example: 90 serum samples or 10 FOF, per air space, is needed to achieve 95% confidence to detect at least 1 sample positive when prevalence is 3% or higher.**

Note the great advantage of family oral fluids (FOF) over serum to detect virus at any given prevalence. There is still the need for a significant number of samples when prevalence approaches near zero. This brings the question about pooling FOF. This has been explored (available next page).

My clients can not afford that sample size. Can I pool aggregated samples such as family oral fluids?

What happens when we pool samples?

Figure 11. A.

When prevalence is low, there will be just a few litters with PRRSv-positive pigs. Target as many litters as possible to achieve higher probability detecting virus. Thus, when the budget is fixed to cover a pre-defined number of PCRs, pooling is a great strategy to sample more crates with the same budget.

- *Example:* Instead of sampling 3 crates running individual PCRs, sample 15 crates and run 3 PCRs in pools of 5.

Figure 11. B.

On the other hand, the greater the intensity of pooling, the more “diluted” the positive crates samples will be with negative crates samples.

Conclusion

Altogether, when considering these two probabilities, pooling is still a great strategy whenever pooling is being done to expand the coverage (number of animal/crates/rooms being included in the sampling).

Figure 11. A. PRRS detection rate and intensity of pooling.

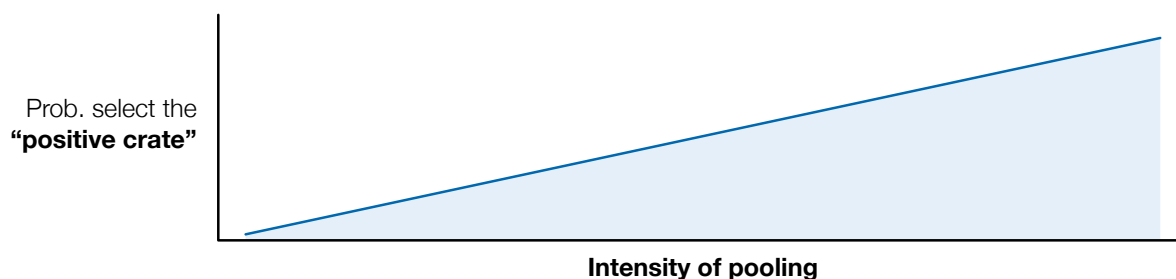
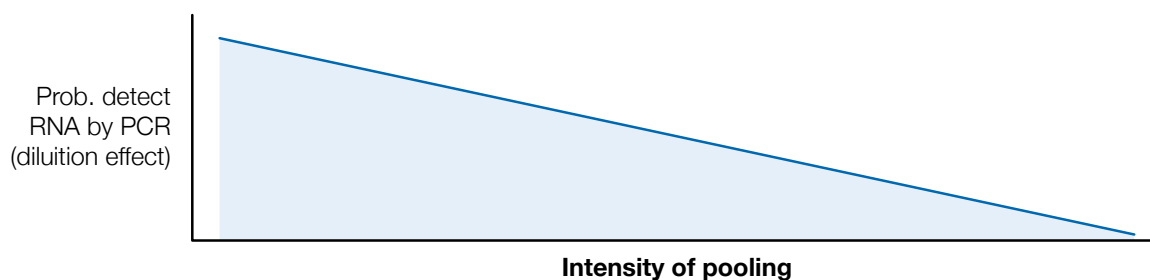


Figure: 11. B. PRRS detection rate and intensity of pooling.



My clients can not afford that sample size. Can I pool aggregated samples such as family oral fluids?

This table provides the probability of sampling **(in black)**, and testing positive by PCR **(in orange)** FOF at different pooling intensities (columns) and crate-level prevalence scenarios (rows).

The probabilities are provided assuming a fixed budget of 3 PCR tests. In other words, 3 tests will be run for PRRSv detection. What is the probability of detecting it at different levels of prevalence and different pooling intensities? The table provides the answers for all combinations of prevalence & pooling level.

It demonstrates that the probability of detecting virus **(orange numbers)** increases as prevalence goes up (5% when there is only 1 positive crate out of 60), and when pooling increases (from no pooling to 1:20).



Osemeke &
Linhares, 2022

Table 6. Probability (%) of sampling at least one PRRSv-positive pen (Number in black) and detecting PRRSv RNA by PCR respectively (in blue) in a 60-crate room given the number of pools submitted using only 3 PCR tests.

Number of litters with positive FOF	3 FOF Individual	9 FOF (3 pools of 3)	15 FOF (3 pools of 5)	30 FOF (3 pools of 10)	60 FOF (3 pools of 20)
1 of 60	5.0 (5.0)	15.0 (14.7)	25.0 (24.10)	50.0 (47.0)	100 (81.0)
2 of 60	9.8 (9.8)	28.0 (27.4)	44.1 (42.7)	75.4 (71.9)	100 (93.3)
3 of 60	14.5 (14.5)	39.4 (38.4)	58.3 (56.9)	88.1 (85.0)	100 (97.1)
4 of 60	19.0 (19.0)	48.8 (47.9)	69.5 (67.7)	94.4 (91.8)	100 (98.2)
5 of 60	23.3 (23.3)	57.0 (56.0)	77.6 (75.9)	97.4 (95.4)	100 (98.7)
10 of 60	42.7 (42.7)	83.1 (82.2)	95.8 (94.9)	100 (99.4)	100 (99.7)
20 of 60	71.1 (71.1)	98.2 (97.9)	99.9 (99.8)	100 (100)	100 (100)

Numbers in black: Probability of sampling at least one PRRSv-positive pen in a 60-crate room given the number of pools submitted using only 3 PCRs.

(Numbers in orange): Probability of detecting PRRSv RNA by PCR in a 60-crate room given the number of pools submitted using only 3 PCR tests.

My clients can not afford that sample size. Can I pool aggregated samples such as family oral fluids?

Both when comparing by week of collection (*Table 7*), or by room (*Table 8*), the agreement of PCR results (positive or negative) of PF and FOF are great, but do not always match.

Reasons include:

- PRRSv transmission may occur between pigs/crates/rooms between PF sampling (2-5 days of age) and FOF (weaning age).
- PF largely represent male piglets (testicle samples), while FOF may not include oral fluids from all pigs within the litter.
- PF represents viremia and virus presence in the pig's body, while FOF is a measuring of virus shedding to the environment.



fieldepi.org



In summary, PF is a practical and accurate method for screening 2-5 days old piglets. Once PCR results start coming back “negative”, there is the need to verify status of weaning-age pigs, which can be done with FOF sampling.

Table 7. Overall agreement in PRRSv RNA detection between processing fluids matched by week of collection. (n= 257 from ~ 135,936 piglets) and family oral fluids samples (n= 2400 from ~ 26,400 piglets).

		Family oral fluids	
		Positive	Negative
Processing fluids	Positive	16 (25.0%)	11 (17.2%)
	Negative	5 (7.8%)	32 (50%)
General agreement: 75%			

Table 8. Overall agreement in PRRSv RNA detection between processing fluids matched by room collected. (n = 114 from ~ 55,776 piglets) and family oral fluids samples (n= 2210 from ~ 24,310 piglets).

		Family oral fluids	
		Positive	Negative
Processing fluids	Positive	18 (15.7%)	14 (12.2%)
	Negative	9 (7.8%)	74 (64.3%)
General agreement: 80%			

1.1.5 Tongue tip fluids (TTF)

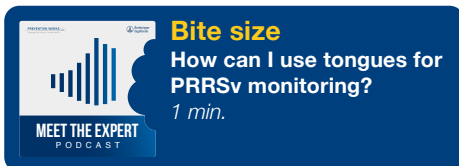
Age group: Dead pigs of any age.

Pro practical, easy, cheap, good quality sample to sequence (clean and low CT values), alternative in farms where castration isn't a practice, non invasive, welfare friendly, very targeted sample.

Caution Still "individual"-based monitoring, but enough samples can be obtained due to stillborn and pre-weaning mortality. More practical, but slightly lower sensitivity compared to matched serum PCR. Freezer needed on farm to store samples.

Action items

- **Positive PCR** = evidence of virus circulation in the age group sampled. If stillborn piglets positive, evidence of vertical transmission as a result of gestating sow virus circulation.



Picture provided by Dr. Isadora Machado.

1.1.5 Tongue tip fluids (TTF)

Equipment



Scissors



Forceps or Pliers



Disposable plastic bags



Disposable gloves

Procedure

Collect tongue tips from dead piglets separated into categories such as:

- x1 bag with tongue tips from **stillbirth**.
- x1 bag with tongue tips from **newborns** (less than 24 hours after birth).

1. Wash and disinfect the material before each sample collection and change gloves.
2. Cut 1 to 2 centimetres (1 inch) of tongue tips with the help of forceps and scissors (E).
3. Place the tongue tips in a disposable plastic bag (F).
4. Quantity of tongue tips per bag: not less than 20 to collect enough fluids.
5. Store the bag in a freezer under -20°C (68°F).



Courtesy: Dr. Isadora Machado.

1.1.5 Tongue tip fluids (TTF)

Sampling technique

STEP 1

Take the tongue out of the mouth.



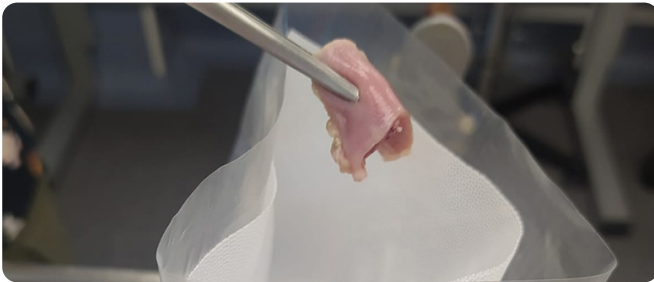
STEP 2

Cut a tip about 2-3 cm with a clean scissors or scalpel.



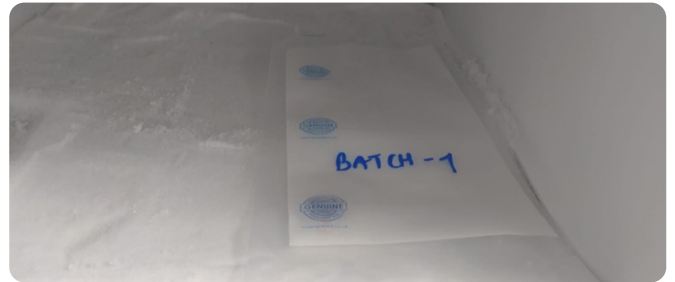
STEP 3

Put the tips in a bag based on batch, site, date, age, etc. (up to 100 tongues per bag).



STEP 4

Store the bag with the tongue tip in frozen conditions.



STEP 5

Add the tips in the bag until the end of the collection period.



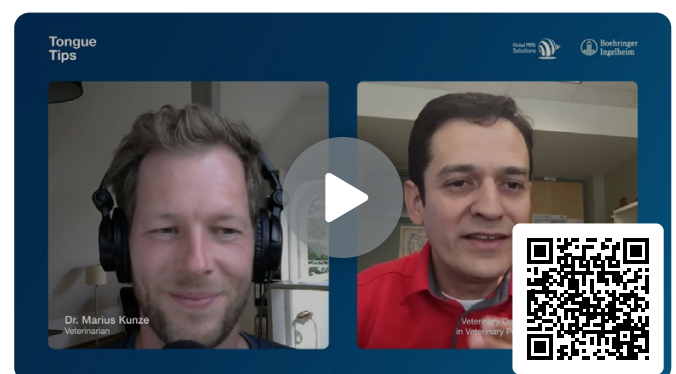
STEP 6

Thaw the bag with the tongue tips and you will obtain a liquid in the top of the bag which can be collected with a syringe.



STEP 7

Send to the lab this liquid sample from tongue tips.



Courtesy: Jordi Baliellas and Dr. Isadora Machado.

1.1.5 Tongue tip fluids (TTF)

Isadora Machado *et al.*, 2022

Table 9. PRRS RNA detection by sample type and age group in three commercial breed-to-wean farms.

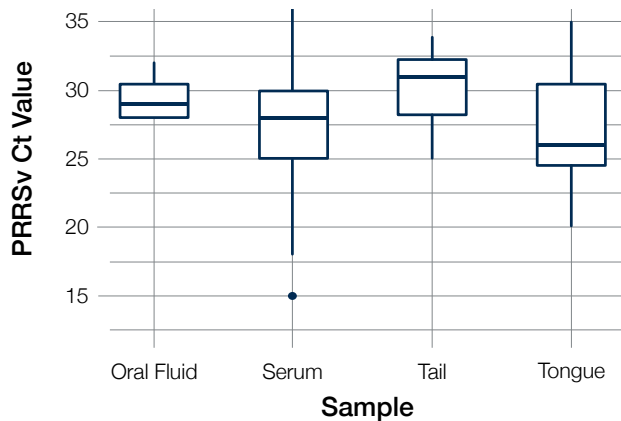
Farm	Age group	Serum		Tongue tips		Processing Fluids		Family Oral Fluids	
		Number of samples and percentage positive†	Ct average (min-max)	Number of samples and percentage positive†	Ct average (min-max)	Number of samples and percentage positive†	Ct average (min-max)	Number of samples and percentage positive†	Ct average (min-max)
A	Newborn	3/9 (33.3%)	34 (32.6 - 36.2)	2/2 (100%)	29.3 (24 - 34.6)	NA	NA	NA	NA
	Processing	9/17 (53%)	24.4 (16.9 - 35.8)	5/5 (100%)	27.8 (24.2 - 31.6)	3/3 (100%)	32.2 (31.3 - 33.8)	NA	NA
	Weaning	4/12 (33.3%)	33.5 (31.2 - 36.9)	2/2 (100%)	36.3 (35.7 - 36.9)	NA	NA	2/17 (11.7%)	34.8 (34.7 - 35)
B	Newborn	3/13 (23%)	26.8 (22.6 - 29.6)	10/11 (90.9%)	26.4 (21.4 - 34.7)	NA	NA	NA	NA
	Processing	2/8 (25%)	19.3 (19.1 - 19.5)	4/4 (100%)	27.8 (20.3 - 35.8)	1/2 (50%)	21.9	NA	NA
	Weaning	3/8 (37.5%)	21.6 (19.0 - 25.0)	6/6 (100%)	29.3 (23.5 - 33.0)	NA	NA	8/35 (22.8%)	32.4 (25.1 - 36.8)
C	Newborn	0/11 (0%)	–	0/5 (0%)	–	NA	NA	NA	NA
	Processing	0/8 (0%)	–	0/7 (0%)	–	0/4 (0%)	–	NA	NA
	Weaning	6/10 (60%)	34.8 (32.3 - 35.5)	0/6 (0%)	–	NA	NA	10/23 (43.4%)	33.1 (30.7 - 36.3)

† Number positive/total pools tested (percentage positive)/NA = not applicable.

PRRSv RNA was generally detected in tongue tips in all age groups in which PRRSv RNA was also detected in serum samples. Further, no PRRSv RNA was detected in tongue tips when serum samples from the same group tested PRRSv RNA-negative. Taken together, these results suggested the potential diagnostic value of tongue tips. Interestingly, the average Ct value from tongue tips was numerically lower than the average Ct value from serum samples in newborns and might be explained by the fact that tongue tips were derived from dead animals, which were potentially more likely to harbor PRRSv.

Is the quality sufficient for sequencing?

Figure 12. Comparison of viral load in different specimens.



For further molecular testing such as sequencing, the quality of the sample (low ct values = high viral load) are crucial. This chart compares the ct values of oral fluids, Serum, Tail fluids and tongue tip fluids. Compared to the other specimen, tongue tip samples had numerically the lowest ct values, close to statistical significance ($p=0.06$).

Reliable sequencing results (ORF5) can be obtained, when Cts are below 32. For whole-genome sequencing Cts should be below 28.

Figure 13. A. Quality chart, oral fluids.

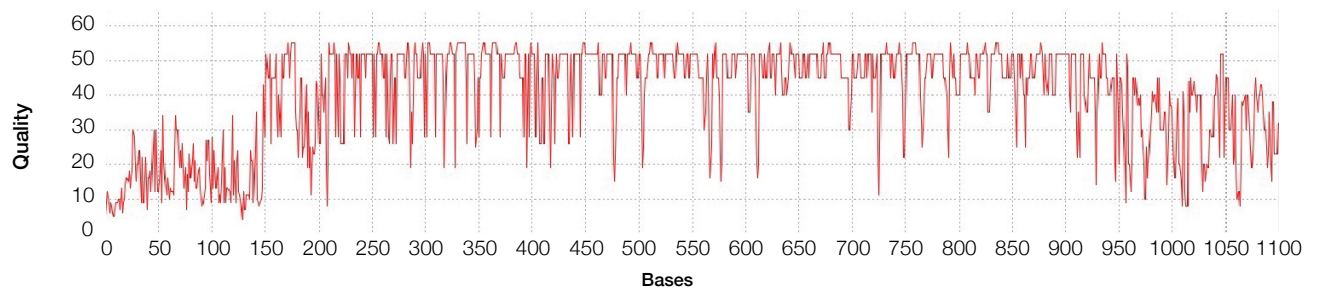
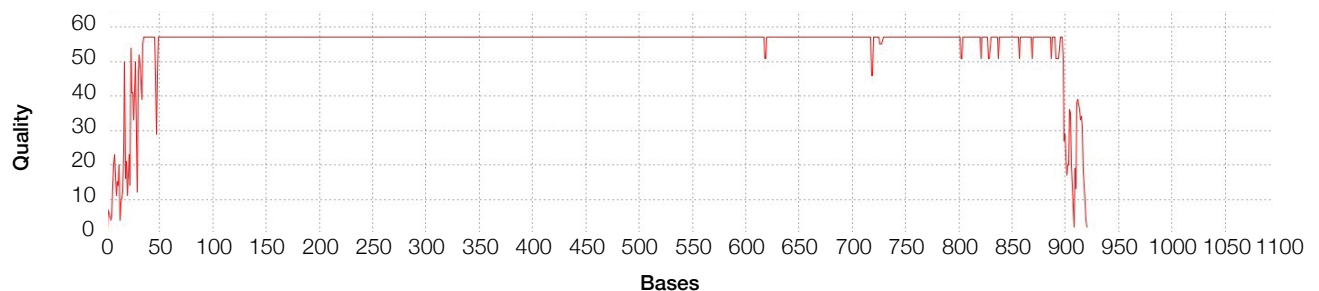


Figure: 13. B. Quality chart, tongue tip samples.



Beside high viral loads (low ct values), good sample quality is needed to obtain reproducible sequencing results. This chart compares oral fluids and tongues in terms of sample quality, showing a consistent higher quality in tongues than in oral fluids.



Baliellas *et al.*, 2019

1.1.5 Tongue tip fluids (TTF)

This farm used tongue tip sampling to monitor their status after breaking with PRRSv. Consecutive negative results (green column) could be reached at week 32 after the outbreak.

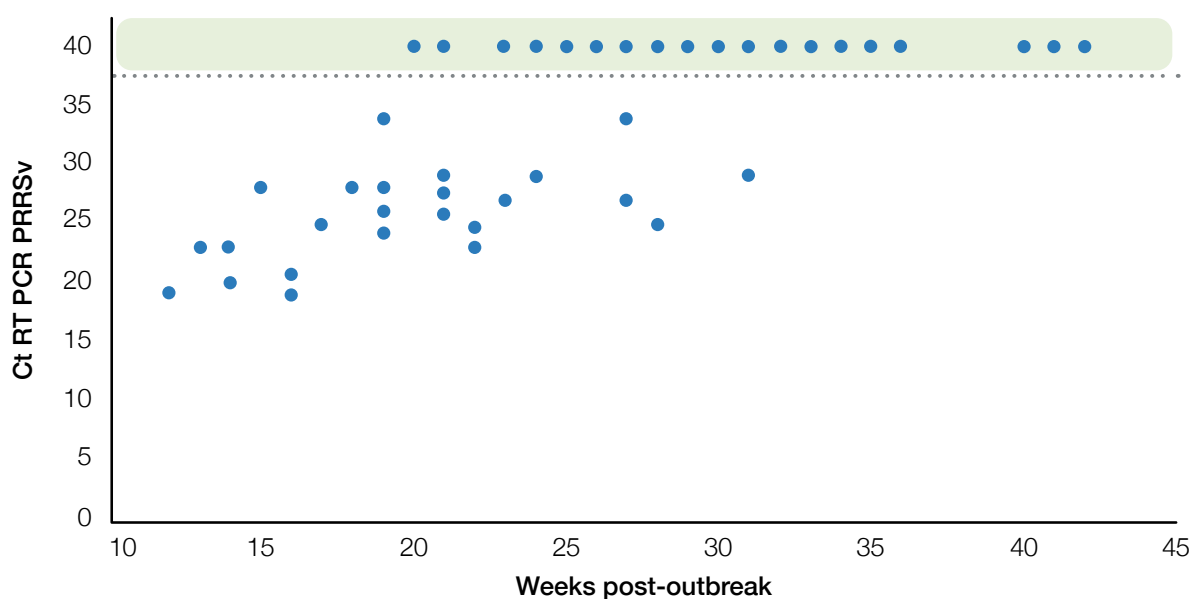
Collecting tongue tips from different age groups (suckling piglets and nursery) helps to understand potential differences in prevalence dependent on the age of the pig. This information is helpful to improve the biosecurity and management practices at different production stages and to consider if we have to introduce new actions to control the disease in case we are not close to consecutive negative PCR results after 30-40 weeks of the outbreak.

Monitoring the outcome of interventions is a critical step in PRRS control. For more information about Step 5 of systematic PRRS control, go to section 3 (page 66).



Baliellas *et al.* 2021

Figure 14. Evaluation of PRRSv stability at birth using tongue tips of stillborn piglets.



Courtesy: Jordi Baliellas.

1.1.5 Tongue tip fluids (TTF): Using tongue tips for PRRSv surveillance in a 1200 sow farm

Case report



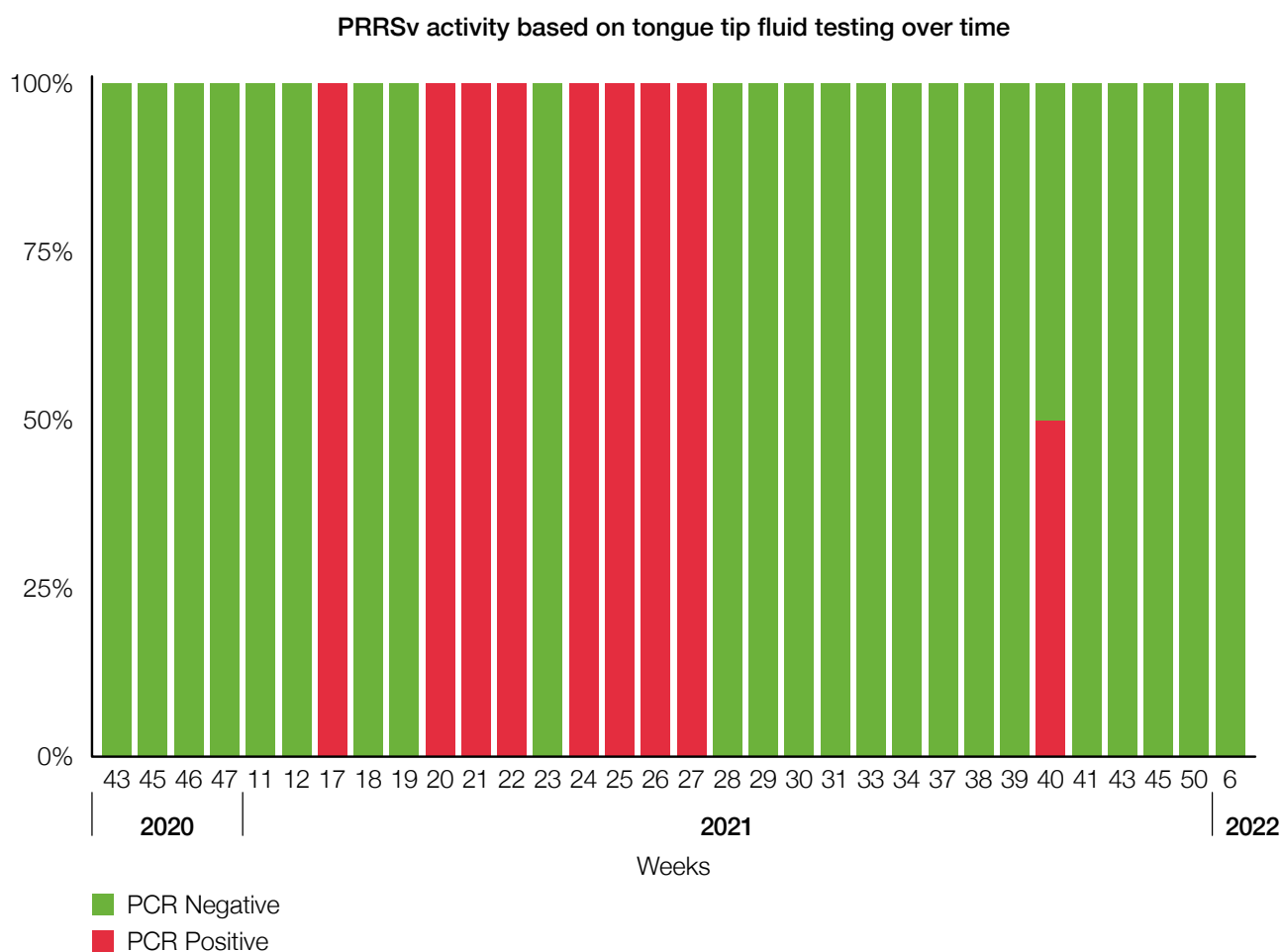
This farm is located in a pig dense area, northeast of Spain. PRRS outbreaks happened frequently in autumn-winter. Historically, for PRRSv Monitoring and Surveillance, 30 due to wean pig were bled.

- At the end of 2020, serum samples were replaced by TTF as are more convenient and cost efficacious sample type.
- After changing to TTF, samples are collected every

farrowing batch by the farmer and send to the laboratory for PCR testing.

- Samples remained negative until February 2021.
- At week 17 (2021) first PRRSv PCR positive samples were detected. During this period, clinical signals were not apparent. Three weeks later (week 20) evident clinical signs were reported with increase of abortions, mummies and dead born piglets.
- Beside changing from weekly farrowing to 2 week batch production (from week 41/2021), the sow vaccine was changed and a mass vaccination approach applied.
- To further reduce virus circulation and protect piglets against detrimental effects of PRRSv, piglets were vaccinated from week 22/2021 onwards.
- 4 weeks after implementation of whole herd vaccination (sows + piglets), an improvement of clinical symptoms was observed, and TTF started to become PRRSv negative again.

Figure 15. Quantity of PRRSv PCR positive and negative Tongue tip fluid samples taken in dead born piglets for respective calendar week.



1.1.6 Tonsil oral scrapings (TOS)

What is TOSc, anyway?

Tonsil-oral scrubbing (TOSc) is a novel sampling technique recently developed for PRRSV surveillance in sows (Li Peng *et al.*, 2024 a, b, c):

- It focuses on rapid, practical sampling without the need for snaring, targeting oral and tonsillar fluids.
- Offers combined sampling of tonsillar exudates and oral fluids for efficient PRRSV RNA detection by PCR.

How does it compare to other sample types?

In field studies performed in endemically infected farms in the US, TOSc outperformed traditional methods (tonsil scraping, serum, oral fluids) in terms of ease and efficiency. The detection rates were similar to that of tonsil, and superior to serum or oral fluids.

The traditional tonsil scraping provides comparable sensitivity but is labor-intensive, while serum and oral fluids have lower detection rates.

Are there tips and tricks on how to sample with TOSc?

Sampling sows in stalls without snaring and focusing on laying sows yielded optimal results.

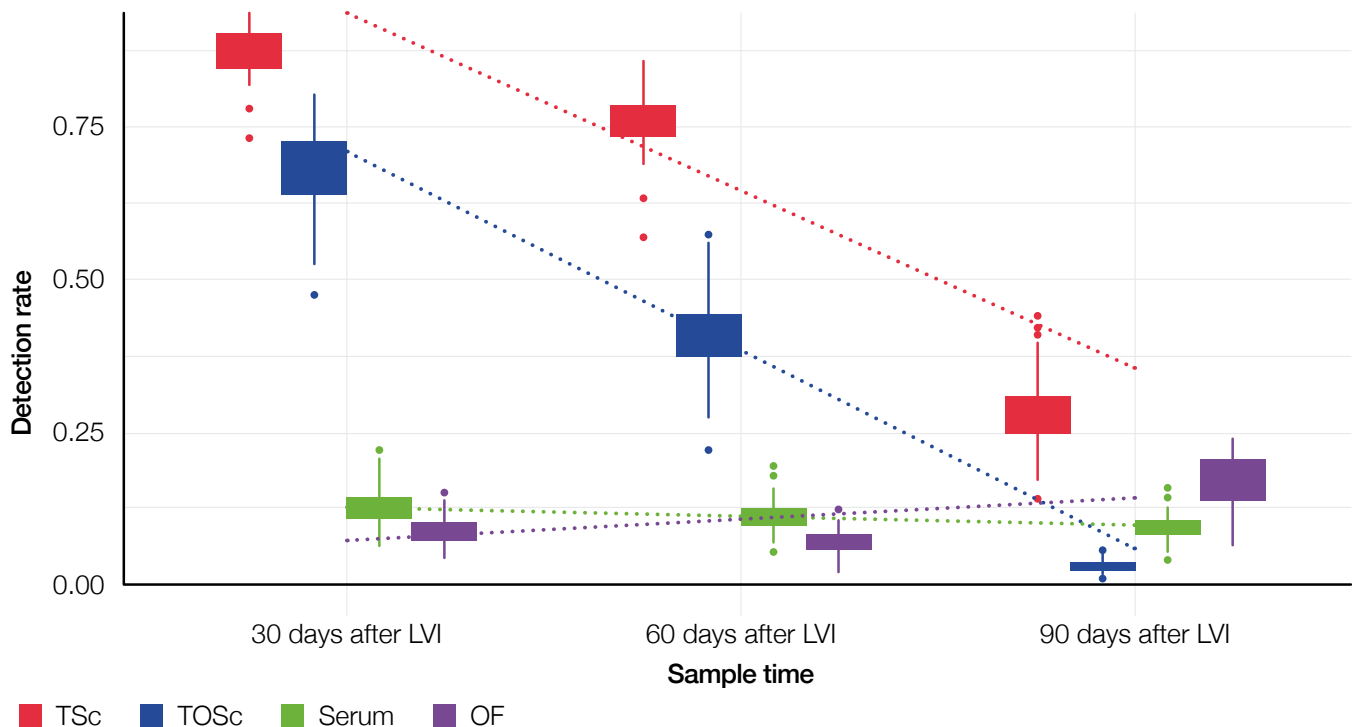
What do we know of PRRSV detection over time using TOSc?

TOSc and tonsil scraping are highly effective early post-infection. As expected both exhibit declining detection rates over time.

When should I consider doing TOSc on my sows?

TOSc is useful when there is a need to understand PRRSV circulation in gestating sows. Common examples of cases include:

- Processing fluids (or tongue fluids) are testing negative for a few weeks. Are the next batches of sows that will farrow the subsequent weeks still shedding or will they likely going to produce negative piglets?
- Stillborn tongue fluids tested PCR-positive this week. Will the subsequent weeks also produce PRRSV-positive pigs at birth?
- I did whole-herd exposure in my herd, and want to verify if my sow population got infected.



Peng Li *et al.*, 2024



Peng Li *et al.*, 2024

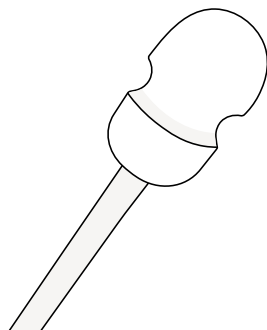


Peng Li *et al.*, 2024

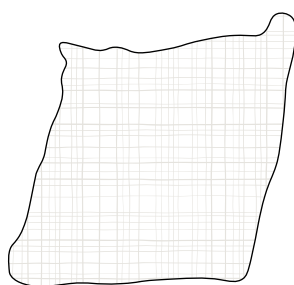
1.1.6 Tonsil oral scrapings (TOS)

How to assemble the tonsil-oral scraping collector (TOSc)

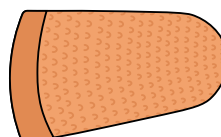
What you need:



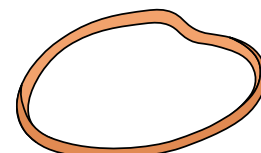
Artificial insemination
(AI) catheter



Cotton gaze



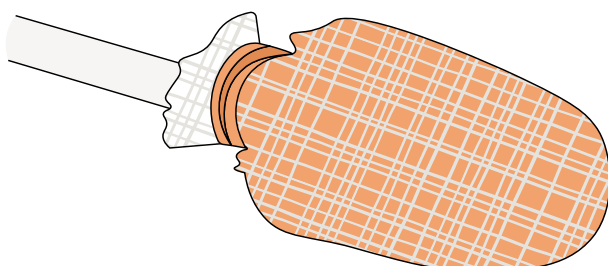
Rubber thimble



Rubber band

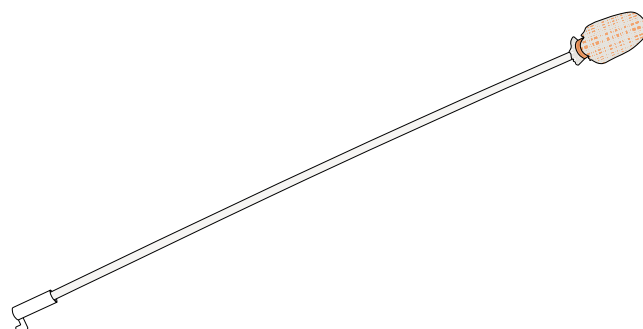
STEP 1

To assemble the TOSc, pull the rubber thimble over the AI catheter head. The thimble will make the tool more abrasive and enhance the sample collection. Wrap around the gaze and fix with the rubber band.



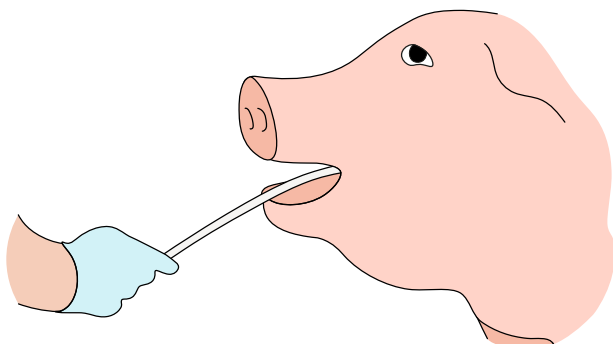
STEP 2

Fully assembled TOSc.



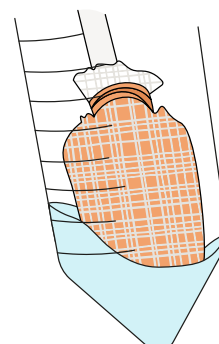
STEP 3

TOSc is collected without restraining the sow, preferably from resting animals, laying down. The head of the AI rod was placed beside the mouth of the sow to attract her attention. The rod was then inserted into the sow's mouth, with the help of a headlamp, directed toward the tonsil area with an upwards angle, and scrubbed back and forth for ten seconds.



STEP 4

The qualified sample was defined as viscous and mucous-like. The head part of the rod with sample on it was cut by a sterilized clip and then transferred to a 50 ml conical tube with three ml of PBS.



1.1.6 Oral swabs

Pro Practical, cheap; good as a method to detect PRRSv at high prevalence; alternative to identify positive pigs from FOF-positive litters.

Caution Still 'individual'-based monitoring. More practical, but lower sensitivity compared to matched serum PCR. Not used for serology.

Action items

- **Positive PCR** = evidence of virus circulation in the age group sampled.



Osemeke *et al.*, 2022



Picture provided by Nathan Vankley.

1.1.7 Air collectors

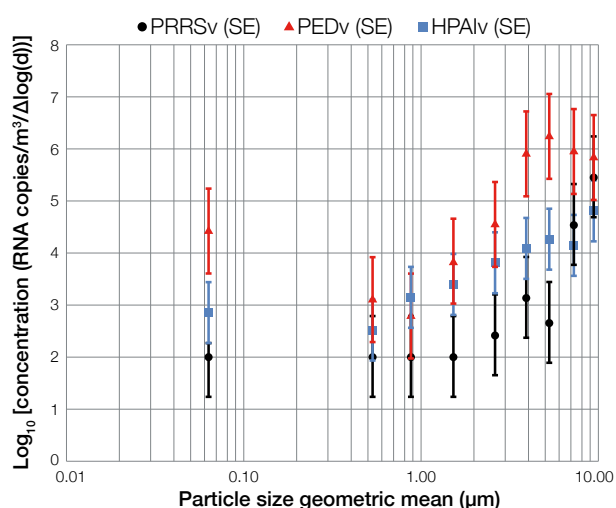
Age group: All ages.

Pro Good evidence of environmental presence of viruses in the air. Good correlation with OF (larger pooled sample). Viral quantitative analyses of total viral load (i.e. qPCR) as well as culture is often used to determine and to quantify virus presence and infectivity.

Caution The detection of viruses in air samples depends on the type of aerosol generated (i.e. pathogen dependent), the sampling and the analytical methodologies used. Certain sampling methods (i.e impaction, desiccation, filtration) as well as airflow, sampling time, and weather conditions (i.e. UV light, temperature and RH%) could affect viral detection and infectivity of pathogens in the air.

Action items The key to achieving a successful air sampling outcome is understanding the type of aerosol generated by a specific pathogen and matching it to the most appropriate air sampling method. Targeting air sampling with zones indicated by sound monitoring of respiratory clinical signs increases the chances of pathogen detection and pathogen monitoring.

Figure 16. Example of viral presence in air samples. Distribution of viral RNA concentration in the air by particle size as detected by the Andersen cascade impactor from aerosols generated by infected animals inside infected premises.



PRRSv: Porcine Reproductive and Respiratory Syndrome virus.

PEDv: Porcine Epidemic Diarrhea virus.

HPAIv: Highly Pathogenic Avian Influenza virus.



1.2 How to improve your sampling technique with risk-based sampling

Risk based sampling will increase your chances to select pigs with a higher likelihood for PRRSv. To better understand which samples to take, we first need to answer the Question:

Is PRRS equally distributed (similar prevalence scenarios) between crates and rooms in positive herds?

"Almeida *et al.*, 2021" analysed samples from different farrowing rooms, side-by-side divided by a wall in the same herd, sampled in the same day.

In each room, FOF from weaning-age litters were collected (blue rectangle), and blood samples from all piglets within each litter taken (1 blue square = 1 piglet). PCR was performed on all samples individually to determine the FOF result and the number of viremic pigs in each respective litter.

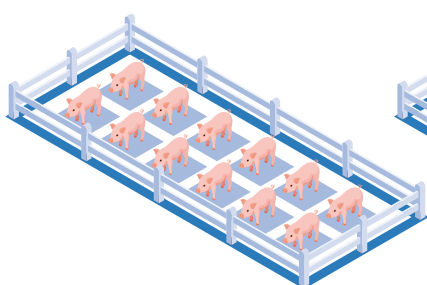
The numbers in the red boxes represent the CT value of PRRSv PCRs. The cut-off to define positivity was set on 37.

| Find the results on the following page.

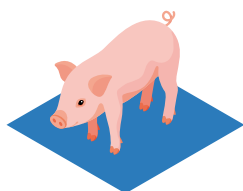


Almeida *et al.*, 2021

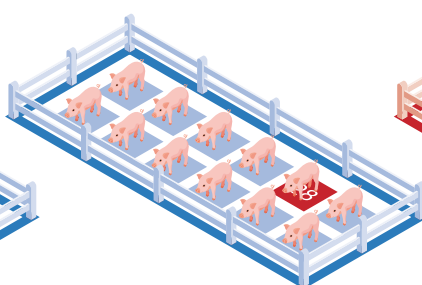
Study design



OF: ⊖ / Serum: ⊖
 PRRSv PCR status of family oral fluids negative.
 Serum samples of each piglet negative.



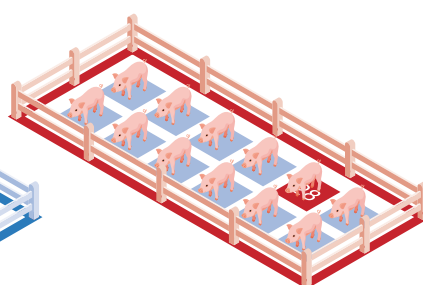
Negative piglet
 (based on serum PRRSv PCRs)



OF: ⊖ / Serum: ⊕
 PRRSv PCR status of family oral fluids negative.
 Single piglet viremic (CT 28).



Positive piglet
 (based on serum PRRSv PCRs)



OF: ⊕ / Serum: ⊖⊕
 PRRSv PCR status of family oral fluids positive.
 Single piglet viremic (CT 28)



CT value of PRRSv PCRs

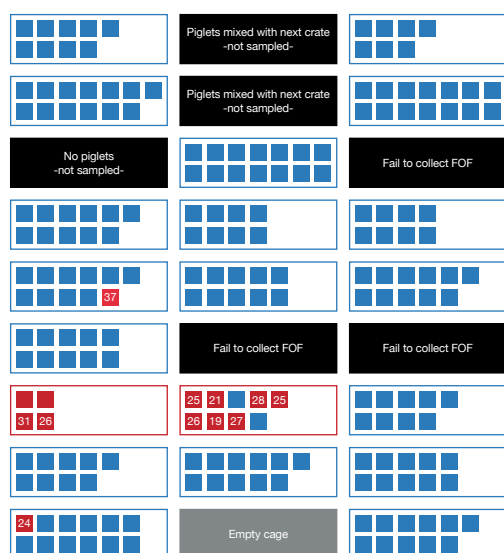
Does PRRSv prevalence change much over time within herds?

Results:

Figure 17. Clustering of PRRS in low prevalence scenarios.

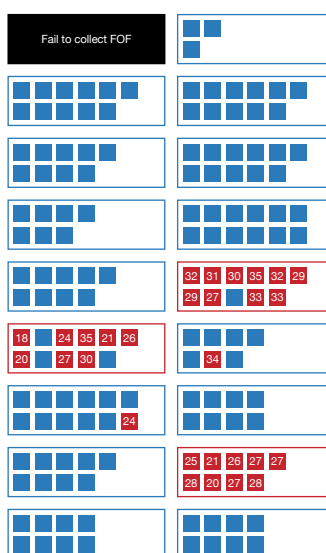
Room A

Piglet prevalence: 6.3%
Litter prevalence: 19.0%
FOF - positive litters = 9.5%



Room B

Piglet prevalence: 19.0%
Litter prevalence: 29.4%
FOF - positive litters = 17.6%



Room C

Piglet prevalence: 57.3%
Litter prevalence: 82.4%
FOF - positive litters = 82.4%



PRRSv status within pigs and crates for 3 rooms* Each rectangle represents a litter, and its color represent the PRRSv PCR status of family oral fluids: blue and red represent negative and positive respectively. The black rectangles represent litters not tested, and gray rectangles represent empty crates. The squares within each rectangle represent the individual piglet status based on serum PRRSv PCR: blue and red represent negative and positive respectively.

Observations:

- Within each litter, there were situations of all pigs negative, all positive, all but one positive, and all but one negative (big variation).
- Piglet prevalence changed significantly between rooms! Caution when extrapolating results from one room to the next.
- Positive crates tended to be clustered (not randomly distributed) when prevalence was low.
- Gilt litters had 4-6 higher odds of testing positive compared to non-gilt litters.

Results support the recommendation of sampling as many pigs, crates, and rooms as possible, and repeating sampling consistently over time.

Particularly in situation of low prevalence, the virus may be located in small clusters (i.e., just a few crates of some rooms), which is easy to miss with low sample size or large interval between sampling events.

Does PRRSv prevalence change much over time within herds?

Conclusion:

The previous figure (*Figure 17*) demonstrated the great variability of PRRSv prevalence within crates. Here there is evidence of variability between rooms of a PRRSv-positive sow farm.

This figure represents PRRSv status at the litter level in 4 rooms of a breed-to-wean herd. Each rectangle represents a litter that FOF collected and tested for PRRSv PCR. Blue and red represent negative and positive PCR result respectively.

There was no evidence of PRRSv in rooms 1 and 2 (all FOFs negative on PCR), and high prevalence on rooms 3 and 4.

Again, demonstrating that PRRSv prevalence may change dramatically room by room. This demonstrates the importance of testing as many crates and rooms as possible. It also reminds the importance of biocontainment/bio-management practices to avoid virus transmission between crates and between rooms.

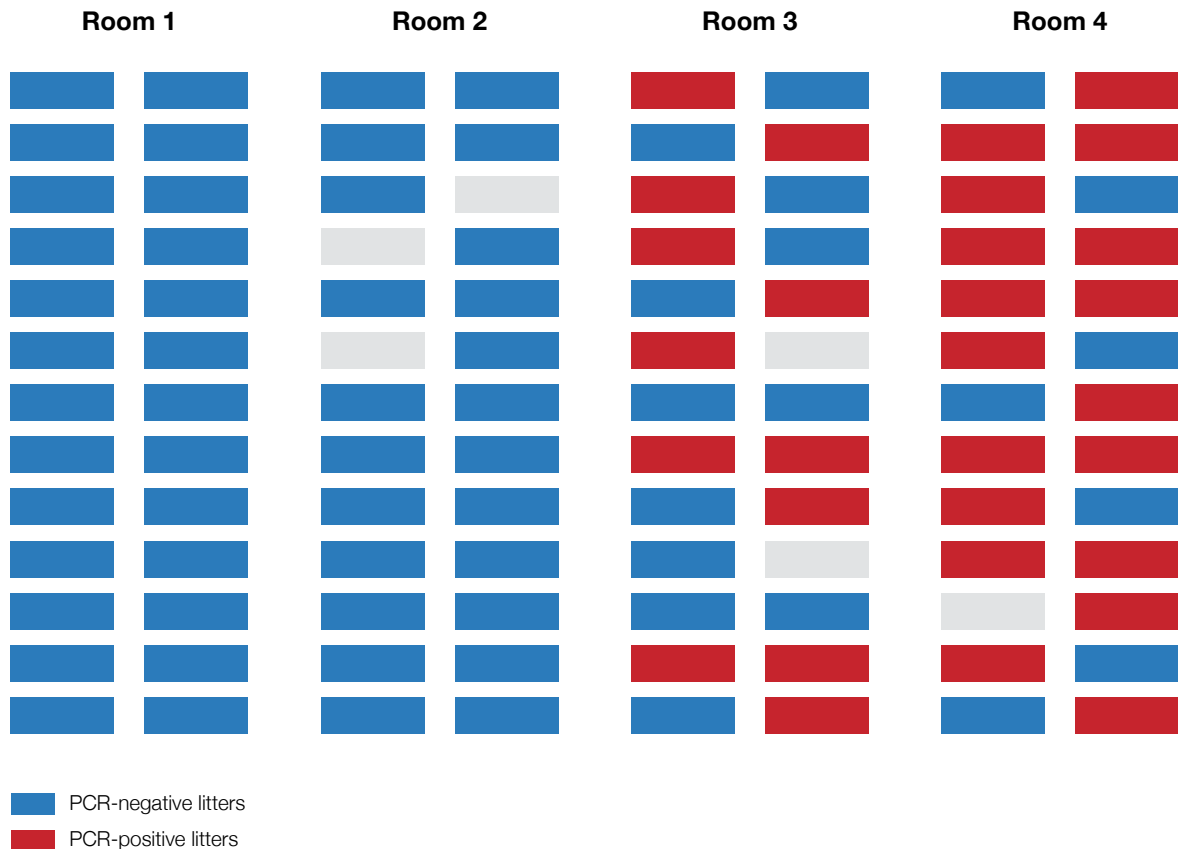


Almeida *et al.*, 2021



Note evidence of plenty of virus circulation in rooms 3 and 4 and lack of any evidence of virus activity in rooms 1 and 2.

Figure 18. Family oral fluids results of 4 farrowing rooms of weaning-age piglets collected in the same farm (same day).



Does PRRSv prevalence change much over time within herds?

Results on previous pages demonstrate significant changes in PRRS prevalence between rooms.

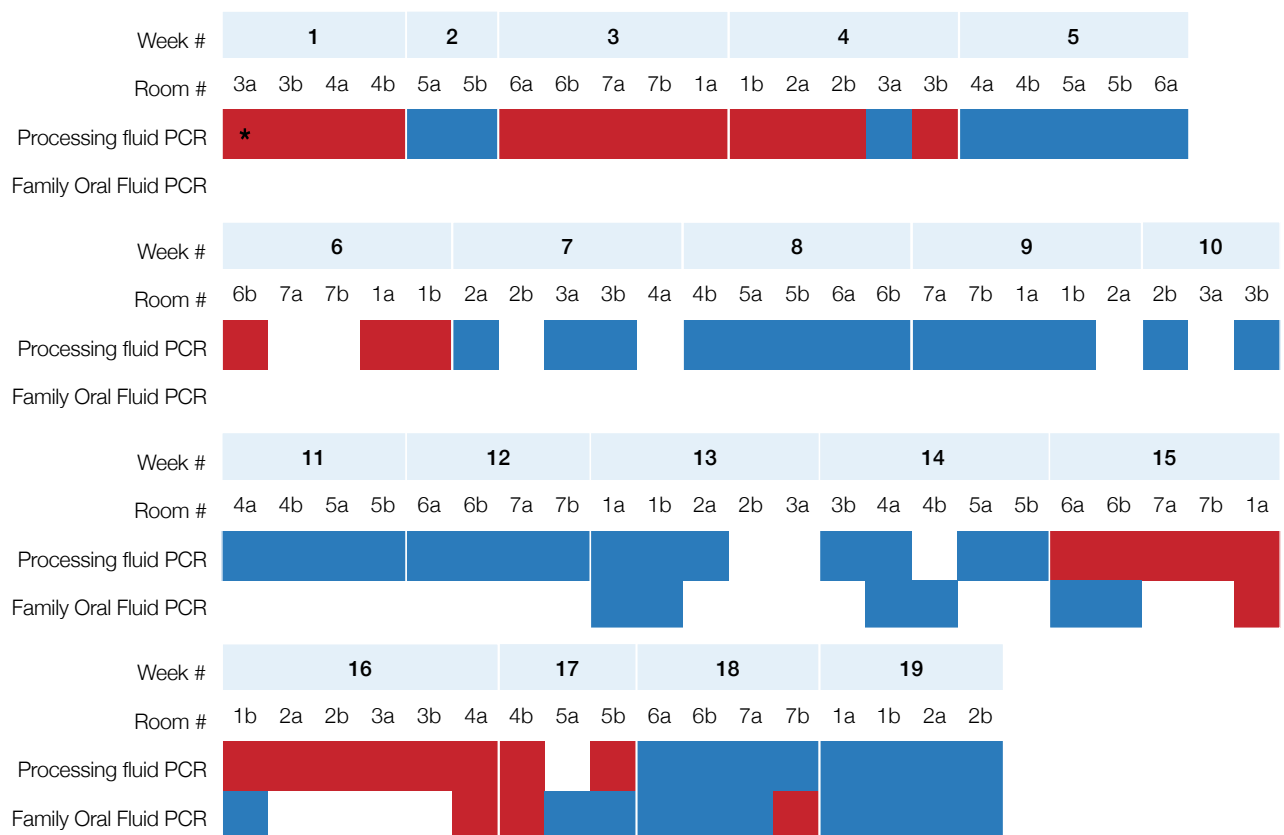
FARM A

- This figure represents PRRSv RNA detection in one farm room after room, week after week.
- The blue and red colours represent PCR result: negative and positive respectively for PF and FOF as indicated in the rows.
- PF were collected from pigs of 3-5 days of age, and FOF collected subsequently in the same rooms when pigs reached weaning-age (18-21 days of age).



Note the intermittent pattern of PRRSv RNA detection by PCR from PF and FOF over time and across rooms.

Figure 19. PRRSv RNA detection (positive, negative) in “Farm A” by: week, farrowing room, and specimen (PF, FOF).



* Each large light blue rectangle represents a week, and each column within rectangle is a room. In **week 1, room 3** and **room 4** were tested positive in Processing fluids.

Does PRRSv prevalence change much over time within herds?

FARM B

Same procedures implemented in farm B as in Farm A. The prevalence seems to be lower, but the intermittent pattern of PRRSv RNA detection is obvious here too.

Conclusion:

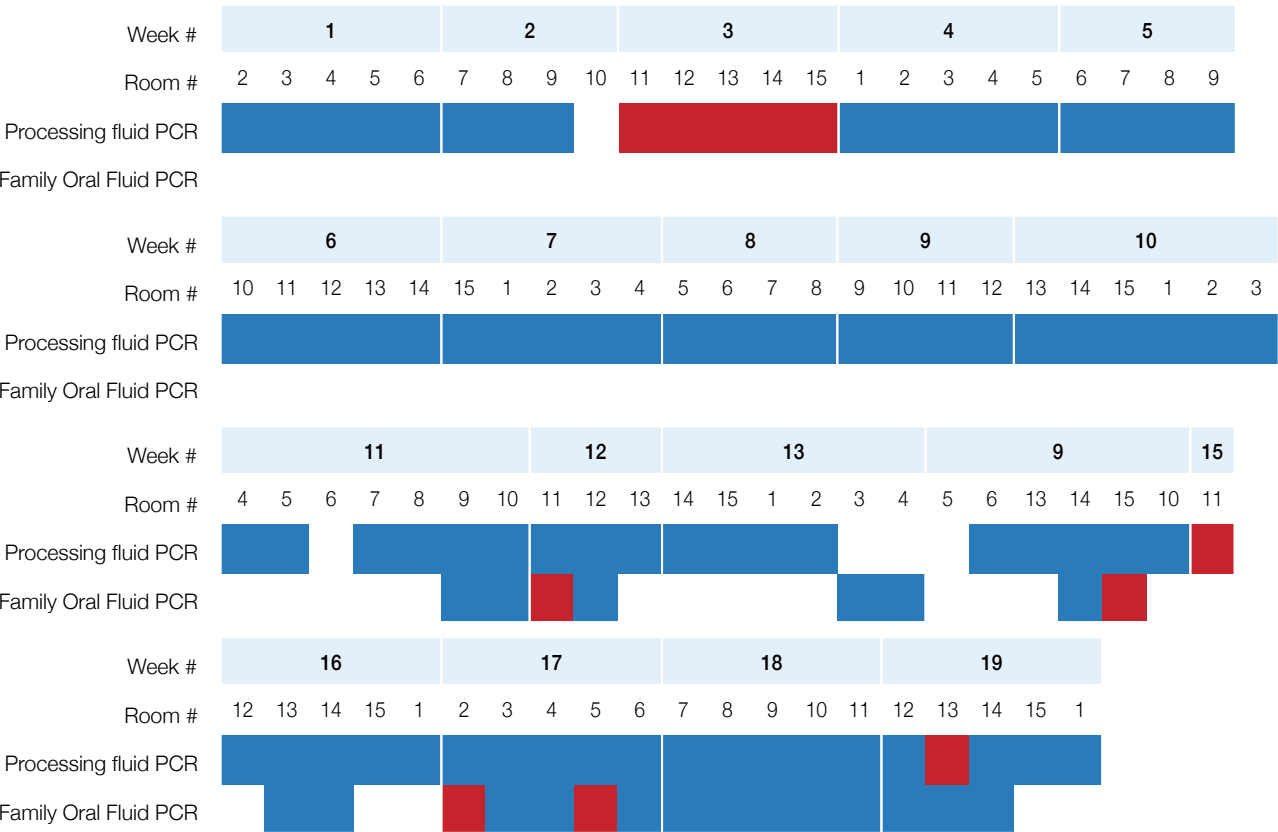
This intermittent pattern of PRRSv RNA detection highlights the importance of sampling as many weeks and rooms as possible, and also demonstrates the importance of internal biosecurity to limit PRRSv transmission across rooms and weeks.

PRRSv prevalence doesnt only change between rooms (space), it also changes over time. Find out more about this "Star Trek Effect" on the following page.



Almeida *et al.*, 2021

Figure 20. PRRSv RNA detection (positive, negative) in “Farm B” by: week, farrowing room, and specimen (PF, FOF).



1.2.1 Star Trek effect: Distribution of PRRSv over time and (geographic) space

PRRSv is not equally distributed (i.e., similar prevalence) between crates and rooms in positive herds. More specifically, there are important changes in prevalence within litters, between crates, between farrowing rooms, and over time (week after week).

This means that MOSS Strategies (Monitoring and Surveillance Strategies) should always consider sampling as many pigs from as many crates and rooms as possible. Also, sampling should be done repeatedly over time, being careful not to extrapolate results from one sampling point to many weeks in the past or future.

PRRSv activity in pig populations is very dynamic, and so should be the MOSS in place to accurately reflect disease status.



How to improve your sampling technique with risk-based sampling?

Clustering of viremic piglets as the prevalence decreases

- Viremic piglets are not randomly distributed in rooms or barns.
- They are in clusters in a few crates.

Litters with increased chance of positives

- Parity 1 litters had 2.82 greater odds of having ≥ 1 piglet PCR-positive.
- Litters < 12 live piglets had 6.13 greater odds of having ≥ 1 piglet PCR-positive.

In practical terms:

- Use zig-zag pattern, sampling pigs/crates/rooms/barns 'spread out' following sampling pattern as close to equidistant as possible (= fixed spatial sampling).
- Give preference to sows parity 1 litters or those with relatively small litters at birth (compared to herd's average).

| Find out more about fixed spacial sampling and zig-zag pattern on next page.



What is fixed spatial sampling?

Spatial sampling consists of drawing samples from the center of the targeted air space (i.e., barn). When multiple samples are taken, those should be spread out equally across the barn, increasing the coverage of the geographic space available, and avoiding oversampling in some areas of the barn while undersampling others. Spatial sampling is better than random sampling when there are clusters of virus presence in the herd.

In other words, when prevalence is low, the positive pigs are likely to be close to each other rather than randomly distributed across crates and rooms.

If there is only one sample being collected, choose a pen in the center of the barn. If two samples are to be collected, select pens with similar distance from the end of the barns and from themselves (i.e., equidistant sampling).

Zig Zag pattern

Same process if sampling multiple pens or barns:
Follow a zig-zag pattern covering as much geographic space in the barn possible, trying to follow an equidistant approach.



Zig-Zag pattern increases the probability of detecting pathogen when at low prevalence clustered in one section of the barn.

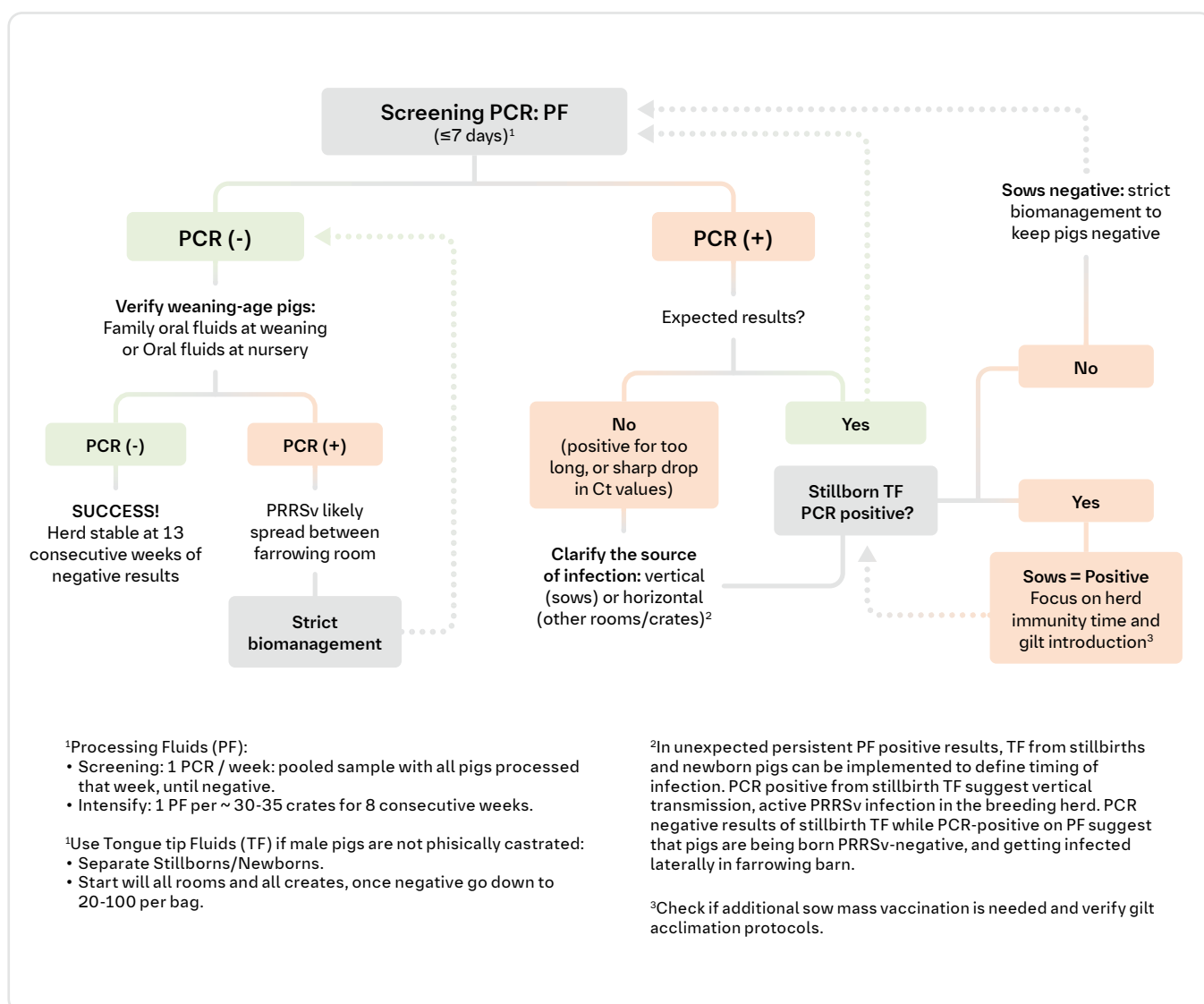
Figure 21. Samples size and distribution per barn.

Barn A		Barn B		Barn C		Barn D	
1	21	1	21	1	21	1	21
2	22	2	22	2	22	2	22
3	23	3	23	3	23	3	23
4	24	4	24	4	24	4	24
5	25	5	25	5	25	5	25
6	26	6	26	6	26	6	26
7	27	7	27	7	27	7	27
8	28	8	28	8	28	8	28
9	29	9	29	9	29	9	29
10	30	10	30	10	30	10	30
11	31	11	31	11	31	11	31
12	32	12	32	12	32	12	32
13	33	13	33	13	33	13	33
14	34	14	34	14	34	14	34
15	35	15	35	15	35	15	35
16	36	16	36	16	36	16	36
17	37	17	37	17	37	17	37
18	38	18	38	18	38	18	38
19	39	19	39	19	39	19	39
20	40	20	40	20	40	20	40

Adapted from J. Zimmerman, (personal communication).

1.3 Sampling guidelines: For herds aiming for PRRS stability

Weaning PRRS negative piglets



1.3 Sampling guidelines: For herds undergoing PRRSv elimination

STEP 1

Start with Processing Fluids (PF):

- **Screening:** 1 PCR/week: pooled sample with all pigs processed that week, until negative.
- **Intensify:** 1 PF per ~30-35 crates for 8 consecutive weeks.

TTF (tongue tip fluids) are great alternative to PF when castration is not done. In this case, collect weekly pooled tongues from stillbirths and neonatal pigs for bags of 20-100 tongue tip samples.

STEP 3

Bring in gilts (i.e., call the herd “Stable”) when:

8 weeks PF-negative, followed by 6-8 weeks PF & weaning-age pigs (14-16 total weeks). Near-zero prevalence for several months.

- Need sample size (~200), or population-based monitoring.
- Intermittent pattern of detection by PCR on PF, FOF (time/space).
- Differentiating between “low” and “zero” prevalence requires aggressive sampling.



When PRRSv is at near zero prevalence, there will be just a couple farrowing crates with viremic pigs. At that time, herd immunity will hide disease. Viremic piglets may be the healthiest in the litter! Breeding herd will be producing pigs at full capacity (low abortions, pre-weaning mortality, and other PRRS-related signs).

When there is unexpected persistent processing fluid-positive results, tongue tips sampling from stillbirths and newborn pigs can be implemented to best define timing of infection.

PCR-positive results from stillbirth tongue tips suggest vertical transmission, meaning that there is active PRRSv infection in the breeding herd.

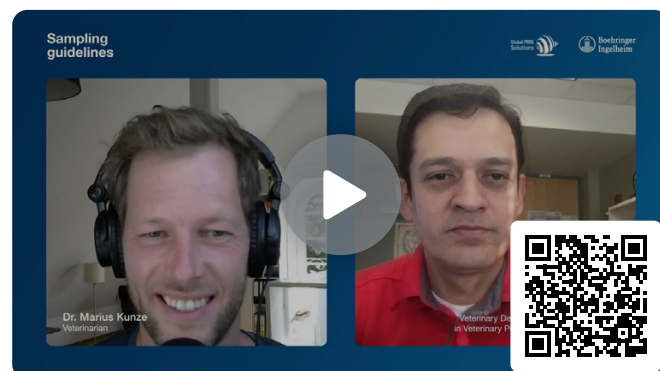
PCR-negative results on a statistical sample of stillbirth tongue tips while PCR-positive on processing fluids and other (older) ages suggest that pigs are being born PRRSv-negative, and getting infected laterally in the farrowing barn due to pigs and people movement across rooms and crates.

STEP 2

Confirm due-to-wean pig status

Need a method to detect < 1-2% prevalence:

- 120 weekly serum samples for 2% prevalence, 210 for 1% prevalence.
- Weekly FOF from all crates (10 crates detected ~2%).
- 6-8 oral fluids in the nursery within 1-2 weeks of weaning (if single source, all in-all out flows).



1.3 Sampling guidelines: For negative herds

To provide evidence of naïve status:

- Antibody testing of PF, FOF, or 30 serum samples from suckling pigs.

To early detect PRRSv introduction:

- Monitor sow feed consumption (Number of gestating sows off-feed).
- Monitor productivity data (Number of abortions, prenatal losses, pre-weaning mortality).
- Weekly testing of PF, representing as many rooms and litters as possible.
- If positive: whole-genome sequence for future epidemiological investigations.
 - *Understand biosecurity gaps and conduct outbreak investigations tracking potential source(s) of transmission.*



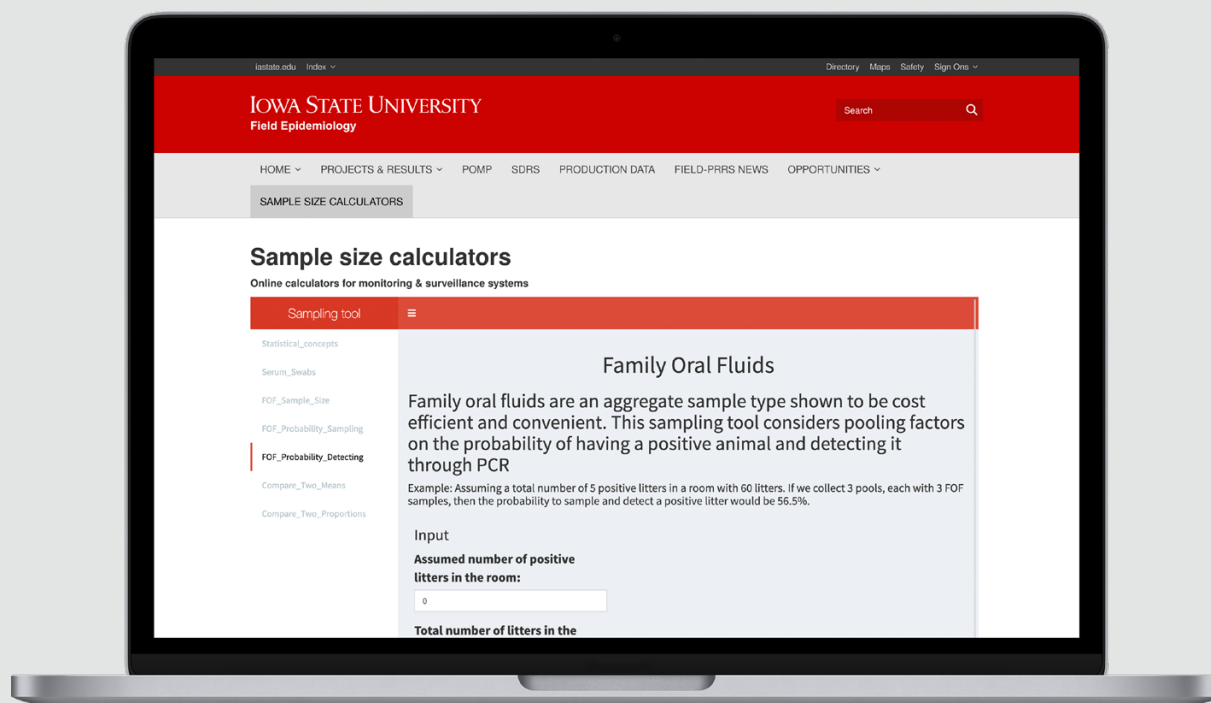
1.4 How to determining sample size for FOF

Calculating the exact number of samples to determine the PRRS status can be easily done with online tool such as the ISU Sample size calculator. It will tell you the number of samples needed to detect PRRS in different prevalences and it helps to validate your current sampling protocol.

Find out at the next page how to use it.



Sample size calculator



1.4 How to determine sample size for FOF

In this example, the budget for sampling consists of 5 PCR tests.

- The sampling **strategy A** consists of collecting 5 FOF samples and testing them individually by PCR.
- The sampling **strategy B** collects 25 FOF samples and test them in pools of 5.

The probability of detection goes from **36% in strategy A**, to **93% in strategy B**, demonstrating the benefit of pooling samples to allow a better coverage of pigs within the sampled unit.



Fieldepi.org

Sampling strategy A: 5 FOFs tested individually

Scenario= 5 “positive” crates out of 60 in a farrowing room.

Input

Assumed number of positive litters in the room:

5

Total number of litters in the room:

60

Pooling factor (number of individual FOF samples in each pool):

1

Number of Pools (Total number of pools collected):

5

Collect 5 FOF and test individually. I.e., 1:1 (no pooling)

Run Calculator

Output

Probability of sampling and detecting a positive litter (%):

(Margin of error +/- 1%)

36.30 %

36% Probability detection

Sampling strategy B: 25 FOFs tested in pools of 5

Scenario= 5 “positive” crates out of 60 in a farrowing room.

Input

Assumed number of positive litters in the room:

5

Total number of litters in the room:

60

Pooling factor (number of individual FOF samples in each pool):

5

Number of Pools (Total number of pools collected):

5

5 pooled PCRs of 5 FOF each. I.e., collect 25 FOF and test in pools of 1:5

Run Calculator

Output

Probability of sampling and detecting a positive litter (%):

(Margin of error +/- 1%)

92.96 %

93% Probability detection

Summary

Accurately monitoring PRRSv activity is key to measure progress and feed decision trees on disease management programs. There are multiple tools available including ongoing, near real-time monitoring of clinical and productivity data. Sharp changes on key indicators are great early predictors of PRRSv activity in swine populations. Diagnostic monitoring can be done with different sample types, including individual pig samples such as serum, blood swab, or tongue tips. Alternatively, veterinarians can employ population-based sampling approaches including processing fluids to screen piglets of 2-5 days of age, family oral fluids on weaning age litters, or oral fluids in pigs after weaning, until the adult age.

Regardless of the sampling approach, it is important to acknowledge that PRRSv circulates in clusters (not randomly) particularly when prevalence is low (i.e. below 20%). For example, the PRRSv-positive piglets will not be equally distributed across crates, rooms, and prevalence will not be constant over time. The PRRSv-positive pigs, instead, will likely be disproportionately located in a few crates of some rooms. What does this all mean? Simply put, it is important that monitoring and surveillance systems consider sampling as many piglets (or litters), crates, and rooms as possible. It is equally important that monitoring should be done repeatedly over time.

It has also been demonstrated that application of pooling of aggregated samples (e.g. Family Oral Fluids) allows a broader coverage of pigs, litters and weeks within a fixed budget. Expanding the coverage of pigs, crates, and weeks, while keeping budget affordable.

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2.0 Classifying PRRSv status: establishing exposure & shedding at the herd level



Why classifying herds for PRRSv?

It is much easier to manage and benchmark what is measured consistently over time following the same standard.

Classifying herds for PRRSv allows:

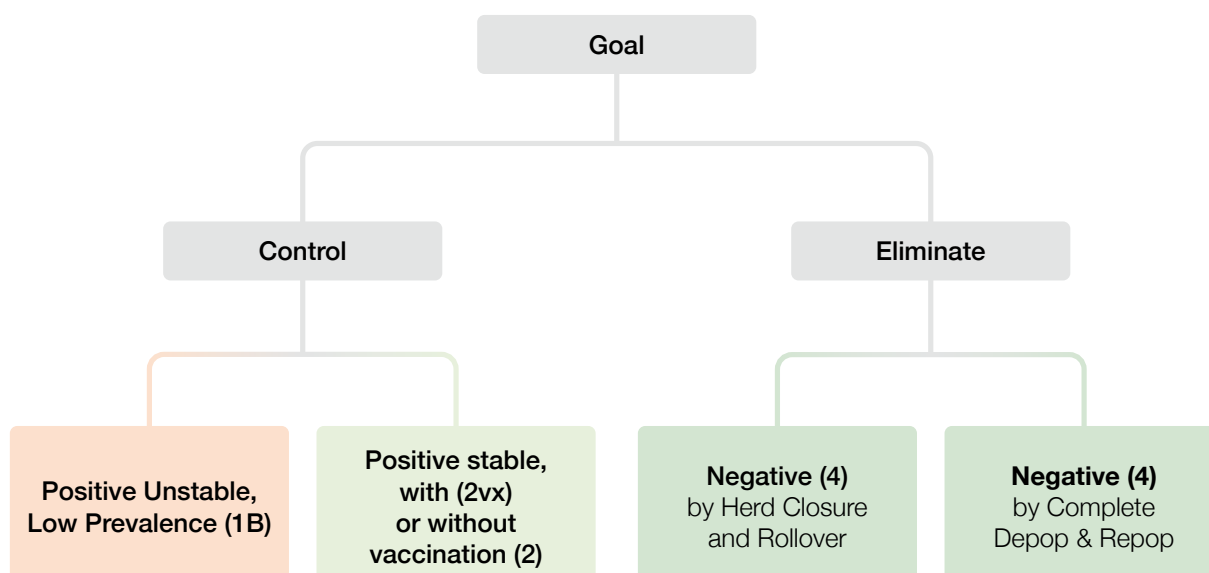
- Providing a roadmap for PRRSv management (i.e., control versus elimination).
- Communication between veterinarians and producers regarding interventions, pig flow, downtimes, and people movement between herds.
- Allows veterinarians and producers to better understand biosecurity by keeping track of the frequency of new outbreaks.
- Conduct epidemiological and economic studies about PRRSv activity and impact in production systems or regions.
- Set premiums and discounts for pigs according to PRRSv status.
- Also, PRRSv status is a great predictor of productivity.



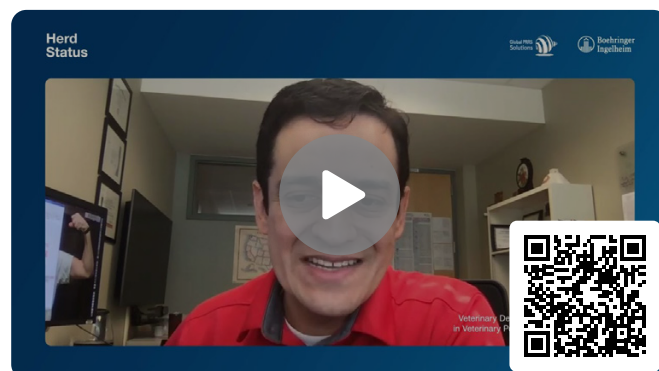
Holtkamp *et al.*, 2022

Figure 22. Updated herd classifications expand the PRRS road map.

The road map for managing PRRSv provides well defined destinations depending on the goal to be achieved.



2.1 AASV classification system for breeding herds















Category	Description	Condition for entry	Condition for stay
1a	Positive unstable High prevalence	Untested/insufficiently tested herds. Outbreak	Same as conditions for entry
1b	Positive unstable Low prevalence	75% of PCR tests for 90 days negative for PRRSv	75% of PCR tests in 90 days negative for PRRSv
2vx	Positive stable w/Ongoing MLV exposure on incoming gilts or sows	wild-type PRRSv negative for 90 days (molecular testing)	PCR tests
2	Positive stable not vaccinating	PRRSv PCR-negative for 90 days	PCR tests
3	Provisional Negative	ELISA negative tests in sentinel gilts, 60 days post entry into the breeding herd	Periodic monitoring (≤ 6 months)
4	PRRSv naïve	ELISA negative tests	Periodic monitoring (≤ 6 months)

2.1 AASV classification system for breeding herds

The new classification system (Holtkamp *et al.*, 2021) has important updates compared to the original system (Holtkamp *et al.*, 2011), including:

- Splitting the positive unstable into the high (1A) and low (1B) prevalence. This reflects the fact that the PRRSv impact on productivity decreases significantly when prevalence decreases (Osemeke *et al.*, 2021).
- Addition of the positive stable with vaccination (2vx) in addition to the plain Stable (2). This accommodates breeding herds focusing PRRSv control rather than elimination, using ongoing exposure of the breeding herd to attenuated virus vaccines. Breeding herds in the 2vx status have lower production impact than truly negative herds upon wild-type PRRSv exposure.
- Also, it was incorporated the use of population-based sampling approaches (processing fluids, family oral fluids), reflecting the new norm of North American herds which incorporated these new sampling approaches due to practicality, lower cost, and higher herd sensitivity compared to serum sampling with 30 or 60 sera per submission.

Holtkamp *et al.*, 2021Osemeke *et al.*, 2021

Herd category	Condition for stay	Shedding status	Exposure status
Positive Unstable, High prevalence (1A) (Shortly after outbreak. Herds that do not meet the criteria for any of the other categories are Category 1A by default).	Same as conditions for entry	 Positive High prevalence	 Positive
Positive Unstable, low prevalence (1B) (When a 90-day period of low prevalence in weaning-age pigs is accomplished, i.e., 75% of PCR-negative results on 90-day period of testing at least monthly).	75% of PCR tests in 90 days negative for PRRSv	 Positive High prevalence	 Positive
Positive Stable (2) (When a 90-day period of sustained lack of viremia due to any PRRSv in weaning-age pigs is achieved).	Monthly negative tests	 Uncertain	 Positive
Positive Stable with vaccination (2vx) (When a 90-day period of sustained lack of viremia due to wild-type PRRSv in weaning-age pigs is achieved in herds using MLV vaccine).	Monthly negative tests	 Uncertain	 Positive
Provisional Negative (3) (Sustained introduction of negative breeding replacements to the PRRSv is required, but some adult breeding animals may still have antibodies to the PRRSv).	Periodic monitoring (≤ 6 months)	 Negative	 Positive
Negative (4) (Starts when there is evidence that the breeding herd is seronegative by ELISA, typically attained after 2 years after status 3, assuming a -50% replacement rate with native gifts).	Periodic monitoring (≤ 6 months)	 Negative	 Negative

2.1.1 Positive Unstable, High prevalence (1A)

Default category for farms that do not test or for farms that had an outbreak. No supporting criteria is needed.



No test



Outbreak



2.1.2 Positive Unstable, Low prevalence (1B)

Conditions to entry

Option 1:



Serum



Pools of 5



Monthly



90-days period

Requirements

75%



RT-PCR negative

Serum from 30 weaning-age piglets tested in pools of five monthly. All batches in a 90-day period are required to be negative by RT-PCR.

Option 2:



Processing fluids



Most litters



Weekly



90-days period

Requirements

75%



RT-PCR negative

One aggregated sample of processing fluids from most litters tested weekly, (or least 4 batches) by RT-PCR. 75% (10 out of 13) of the batches in a 90-day period are required to be negative by RT-PCR.

Option 3:



Family oral fluids
weaning age litters



Litter x ≥20
Pools of 5



Monthly or batch

Conditions to stay



Monthly

Requirements
≥75%
(3 out of 4 tests)

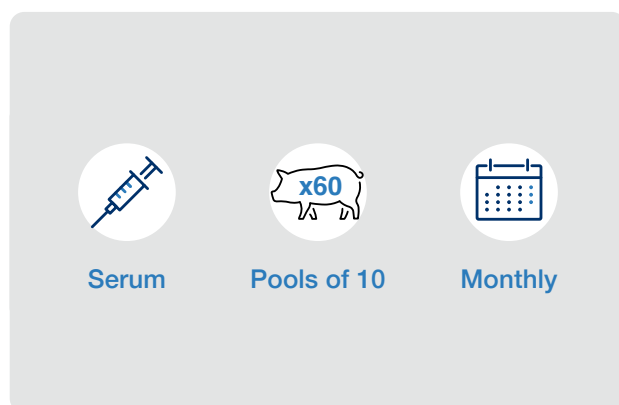


RT-PCR
negative




2.1.3 Positive Stable (2)

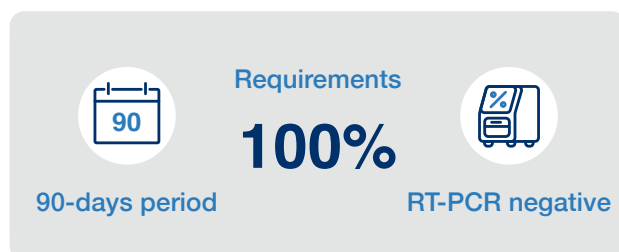
Conditions to entry

Option 1:





Option 1 testing requirements:

-  Serum
-  Pools of 10
-  Monthly

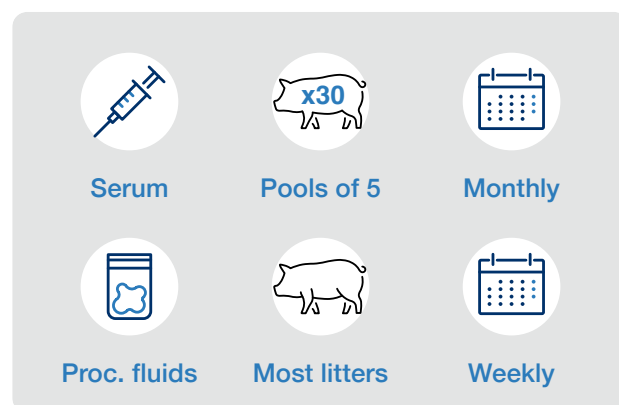


Option 1 requirements summary:







-  90-days period
- 100%** Requirements
-  RT-PCR negative

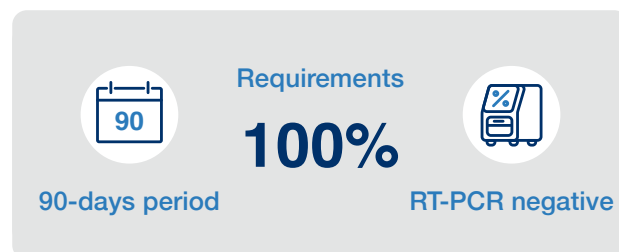
Serum from 60 due-to-wean piglets tested in pools of ten monthly. All batches (at least 4) in a 90-day period are required to be negative by RT-PCR.

Option 2:





Option 2 testing requirements:

-  Serum
-  Pools of 5
-  Monthly
-  Proc. fluids
-  Most litters
-  Weekly

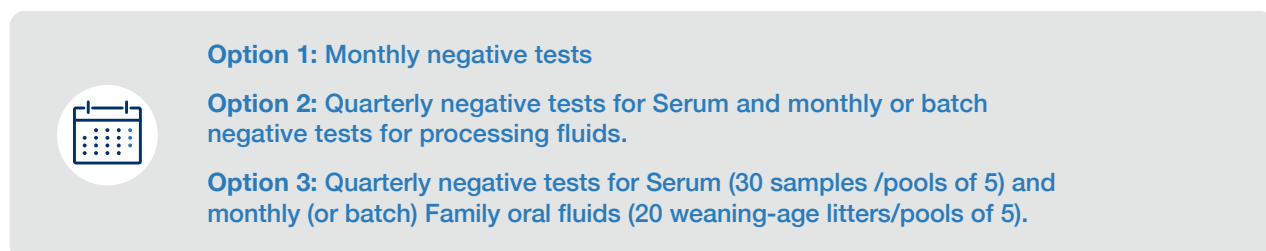


Option 2 requirements summary:

-  90-days period
- 100%** Requirements
-  RT-PCR negative

Serum from 30 due-to-wean piglets tested in pools of five monthly (at least 4) and one aggregated sample of processing fluids tested weekly (at least 4) by RT-PCR. All samples are required to be negative in a 90 day period.

Conditions to stay




Conditions to stay options:

- Option 1: Monthly negative tests**
- Option 2: Quarterly negative tests for Serum and monthly or batch negative tests for processing fluids.**
- Option 3: Quarterly negative tests for Serum (30 samples /pools of 5) and monthly (or batch) Family oral fluids (20 weaning-age litters/pools of 5).**

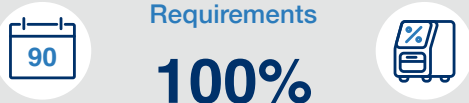
2.1.4 Positive Stable with vaccination (2vx)

Conditions to entry

Option 1:



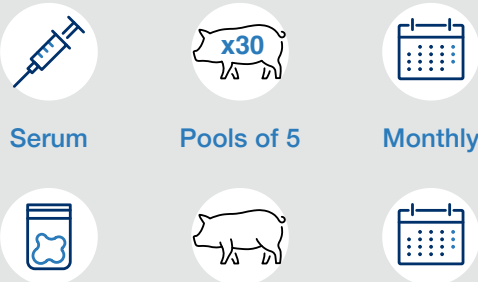
Serum Pools of 10 Monthly




90-days period **Requirements 100%** RT-PCR negative

Serum from 60 due-to-wean piglets tested in pools of ten monthly. All batches in a 90-day period are required to be negative by RT-PCR.

Option 2:




Serum Pools of 5 Monthly
Proc. fluids Most litters Weekly



90-days period **Requirements 100%** RT-PCR negative

Serum from 30 due-to-wean piglets tested in pools of five monthly and one aggregated sample of processing fluids tested weekly by RT-PCR. All samples are required to be negative in a 90d period.

Conditions to stay



Option 1: Monthly negative tests
Option 2: Quarterly negative tests for Serum and monthly or batch negative tests for processing fluids.
Option 3: Quarterly negative tests for Serum (30 samples /pools of 5) and monthly (or batch) Family oral fluids (20 weaning-age litters/pools of 5).

If a positive sample is found more than two weeks after the herd is vaccinated, the presence of a wild type virus should be determined/differentiated by sequencing.

2.1.5 Provisional Negative (3) & Negative (4)

Conditions to entry



Serum



PRRS naive
replacement gilts,
60d after introduction



Once

Requirements



ELISA negative

Provisional Negative (3)

Serum from 60 negative breeding replacements by ELISA 60 days after their initial introduction. Absence of positive results is required to achieve this category.

Negative (4)

Same sampling guidance as for Category 3, but in adult breeding animals. Absence of positive results is required. Repopulated herds with naive animals fall in this category after the tests are negative.

Conditions to stay

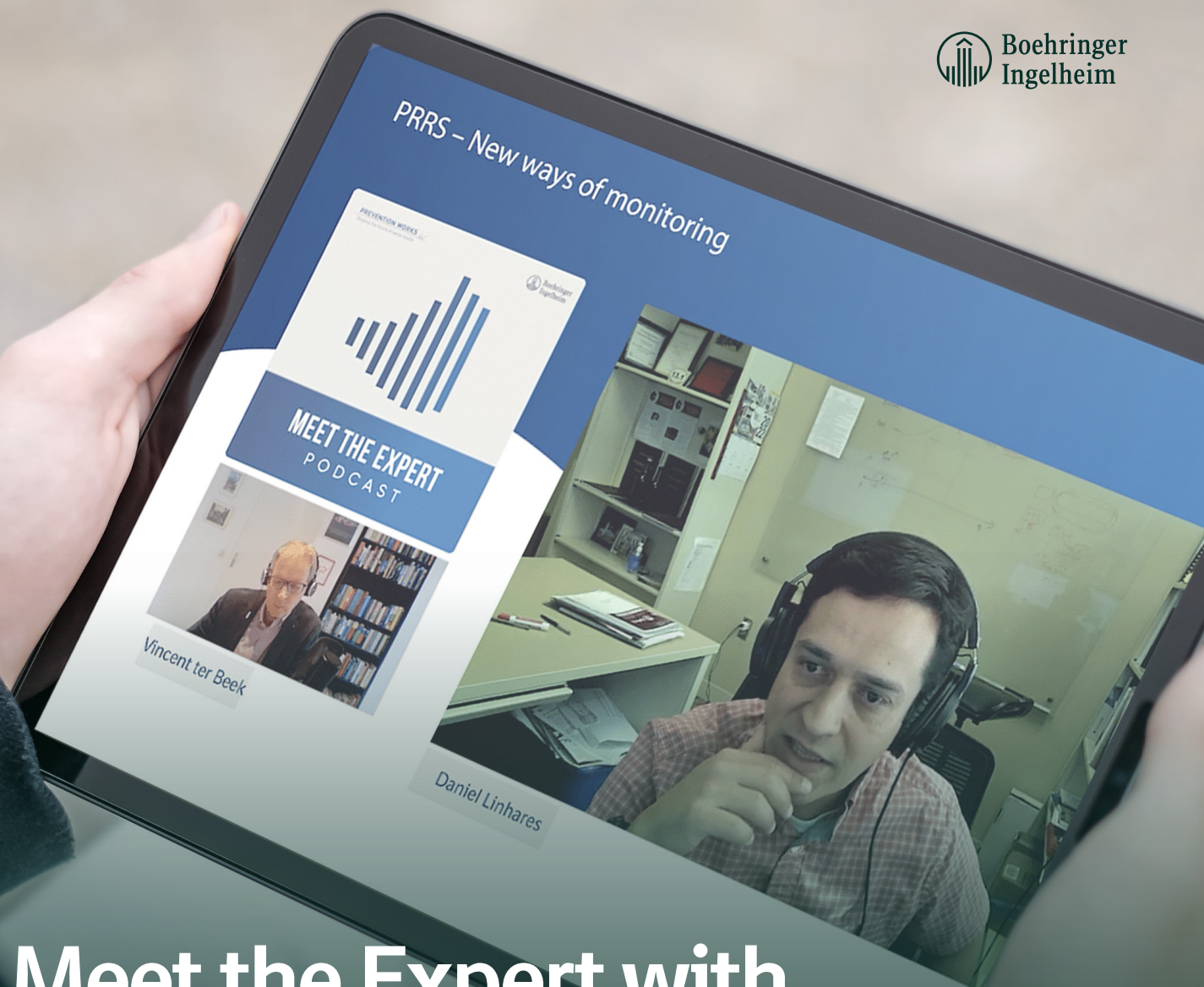


Periodic monitoring
(≤ 6 months)

Summary

- The AASV PRRSv classification scheme can be implemented to herds of different sizes and layout. It allows standardizing the measurement of PRRSv activity, leading to many different applications as outlined in the introduction of this section.
- Under the 2022 PRRSv classification version, veterinarians and producers have different sampling options for monitoring and classifying herds, using serum or alternative population-based monitoring. It also takes into account that not all breeding herds target complete virus elimination (status 4). Instead, some herds may target PRRSv control (status 2 or 2vx), keeping herd immunity which is economically important for herds expecting an outbreak frequency of 2-3 years.





Meet the Expert with Prof. Daniel Linhares

Veterinary Degree and MBA in Brazil
PhD in Veterinary Population Medicine in the US

PRRS - New ways of monitoring

This episode is the first of two podcasts with associate professor Daniel Linhares of Iowa State University in the USA. He is a well-known global expert on Porcine Reproductive and Respiratory Syndrome virus. This first episode revolves around monitoring of the virus in sow farms.

Listen now



3.0 Key factors on PRRSv control: Moving up statuses



Why is it important to control PRRS and move up the status?

Dependent on possibility and regional circumstances, either eradication or control (PRRS stability) has a direct impact on productivity. Derald Holtkamp and team compiled data from the US Department of Agriculture, Swine veterinarians, and production records.

His research clearly indicated the impact of PRRS and the benefit aiming for PRRS negativity (**Status 3 + 4**) or stability (**Status 2**) and moving the virus as far downstream as possible. The graphs below demonstrate the impact of PRRS on breeding herds and on growing piglets.

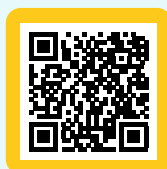
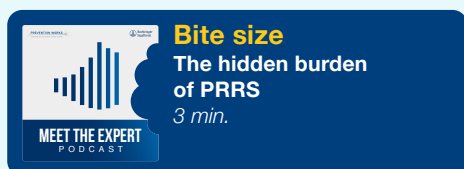


Figure 23. Impact of PRRS status on reproductive performance (sows).



	Current PRRSv infection status			
	PRRSv-infected		PRRSv-free	
	PRRSv outbreak in previous 12 months			
	Yes	No		
	PRRSv infection status just prior to outbreak			
	PRRSv-infected	PRRSv-free		
No. of pigs born alive per litter farrowed	10.6 ± 0.2	11.0 ± 0.1	11.2 ± 0.1	11.6 ± 0.1
Pre-weaning mortality (%)	18.0 ± 1.2	13.7 ± 0.8	12.6 ± 0.8	12.2 ± 0.8
No. of litters per mated female per year	2.33 ± 0.04	2.38 ± 0.04	2.39 ± 0.04	2.45 ± 0.04
Breeding-female culling rate (%)	47.7 ± 2.6	49.8 ± 2.3	50.5 ± 2.2	50.7 ± 2.2
Breeding-female death rate (%)	9.5 ± 0.8	9.6 ± 0.6	9.1 ± 0.5	8.4 ± 0.6

Figure 24. Impact of PRRS status on fattening pigs (growing pigs).



	PRRSv status of pigs at weaning		
	Positive		Negative
	PRRS status at marketing		
	Positive	Positive	Negative
Mortality rate (% of pigs placed)	9.3 ± 0.9	7.4 ± 0.8	6.0 ± 0.8
Average Daily Gain (g/day)	692.1 ± 18.2	695.9 ± 18.0	709.8 ± 18.0
Feed conversion rate (g feed/g gain)	2.57 ± 0.04	2.57 ± 0.04	2.61 ± 0.04
Pigs sold in the primary market (% marketed)	95.8 ± 0.6	95.7 ± 0.6	96.4 ± 0.5

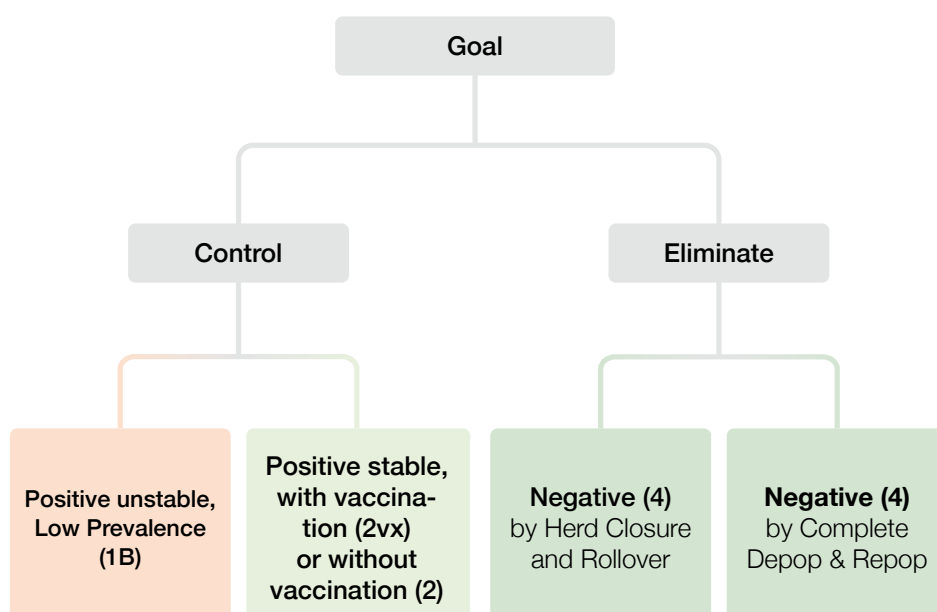
3.1 Benefit of PRRS control

What	Control path	Elimination path
Virus circulation (prevalence)	Low	Zero
Type of PRRS virus	Reduce genetic diversity of PRRSv ideally from wild type to MLV	From wild type to none
Incoming gilts	Previously immunized (2-3 months), non-shedding	Naïve when prevalence reaches zero
Semen	Naïve	Naïve
Weaned pig vaccination strategy	Depends on probability of infection and type/severity of PRRSv in the neighbourhood*	Depends on probability of infection and type/severity of PRRSv in the neighbourhood*

*From multiple doses to no vaccination (negative pigs placed into PRRSv-free region).

Figure 25. Road map for managing PRRSv in breeding herds, with well defined “destinations” depending on the goal.

The first step in controlling PRRS is to identify clear and attainable goals. The goals of a program will either focus on PRRS control or PRRS elimination. To help controlling the Disease in an strategic way, the 5 Step process helps to remain focused and successful. Learn more about this systematic approach on the next pages.



Adapted from: Dr. Derald Holtkamp, 2021

5 Step for systemic PRRS control

PRRS control is much more than just vaccination alone. It requires a comprehensive understanding of the disease and the production system under threat, the use of multiple management tools, and a systematic approach to management.

The 5-step process was developed to help producers and veterinarians on different farms work together, share information and align their activities so that all involved stakeholders are working towards the same goals in PRRSv control.

| Want to know more about the 5-step process? Watch the video!



Download the applied 5 Step worksheet and start controlling PRRS in your system.



5 Step for systemic PRRS control

Step 1. Identify desired goals

The first step in the process is for the producer and veterinarian to identify clear and attainable goals. The goals of a program may vary significantly but will ultimately be focused on one of the following key objectives: **PRRS control** or **PRRS elimination**.

If Control, establish the desired final destination (low prevalence, plain stable, or stable vaccinated). For grow-finish herds, the definition of success may be to decrease the pressure of wild-type infection and improve growing performance (survivability, growth rate, feed efficiency). For regional controls, the goal may be to decrease PRRSv incidence and prevalence.

Step 2. Determine current PRRS status

PRRS status is determined through the evaluation of PRRSv shedding and PRRSv exposure.

Step 3. Understand current constraints

As an **important pillar** of the 5-Step Process, COMBAT enables us to understand current constraints by analyse, visualize, benchmark, and guide to improve biosecurity and management in farms.



Step 4. Develop solution options

The farmer and veterinarian should work together to develop solution options, including biosecurity, pig flow and vaccination programs. The solution options are usually as specific as the constraints in the farm. Free online tool (e.g. COMBAT) help to identify and suggest individual solutions.

Check out the next section “Moving up statuses” (page 70) to get general recommendation on how to climb the ladder and improve your PRRS status.

Step 5. Implement and monitor preferred solutions

Implementation of a PRRS control program should be an active communication process involving all individuals who will be affected by the plan or potential change.

Vets and producers should align on performance parameters that will help to determine the success of the PRRS control plan. These key performance indicators (e.g. PRRSv status at weaning, close-out performance, clinical observations, Diagnostics, TTS, TTBP, Mortality, ADG, Antibiotic use) may vary and are dependent on the desired goal (Step 1). With this information gathered, a sense on how well a program is working can be obtained.

5 Step for systemic PRRS control

Step 5. Implement and monitor preferred solutions

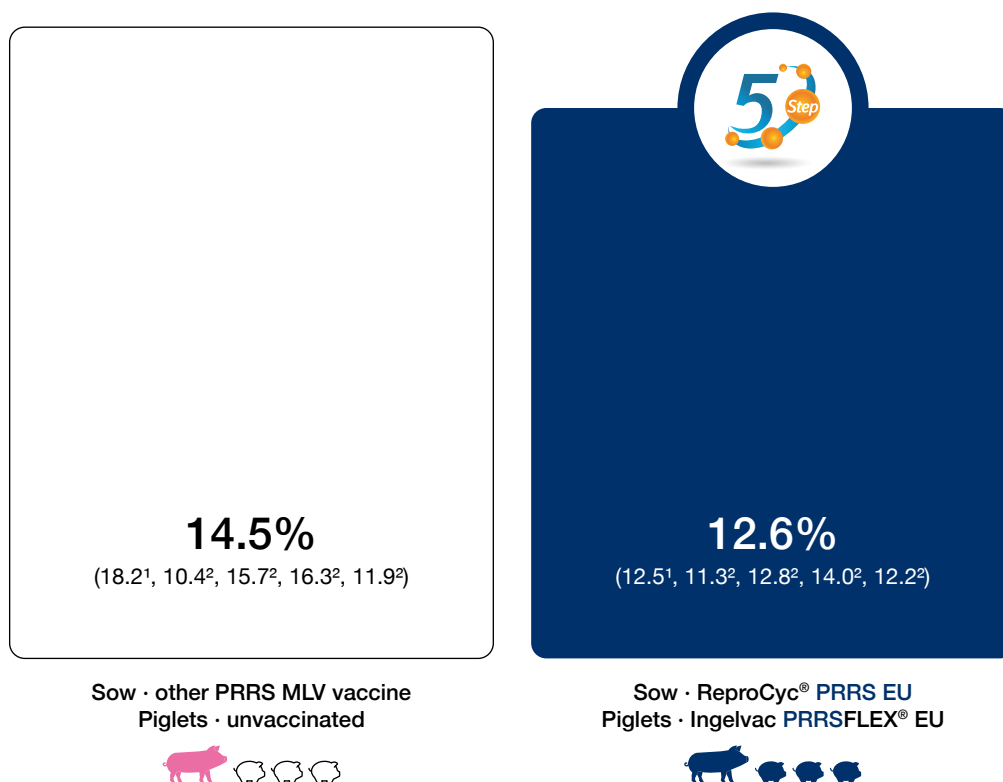
Goal: Improving performance parameter and reduce pre-weaning mortality.

Case report: An important Objective for these endemical infected sow farms, located in Germany and France, was to improve performance parameter such as pre-weaning mortality. Beside an existing sow vaccination program, implementing piglet vaccination with PRRS FLEX EU and improving Management practice, helped to reach the achieved target.

Monitoring the efficacy of implemented solutions by specific performance parameter helps to determine the success of the 5 Step program.



Figure 26. Step 5, monitors the outcome of PRRS control program by tracking performance indicators such as pre-weaning mortality.



Hoffschulte¹ and Kunze
2018, ESPHM



Messenger² et al.,
2019, ESPHM

5 Step for systemic PRRS control

Step 5. Implement and monitor preferred solutions

Goal: reach time to baseline production (TTBP) as soon as possible.

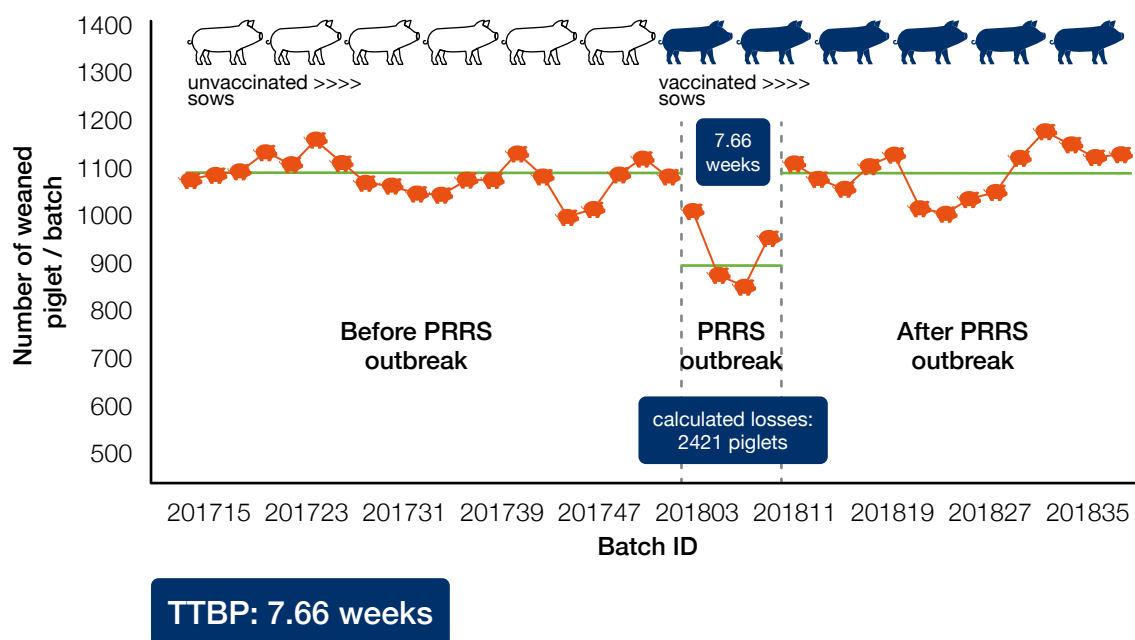
TTBP: time in weeks, to reach the same Number of weaned piglets as before the PRRS outbreak.

After breaking with PRRSv, the objective of this 1000 sows farm was to achieve baseline production as soon as possible to mitigate the negative effects. To control the disease mass vaccination, associated with **strict internal biosecurity measures** were introduced.

7.66 weeks later, productivity reached in average the same level as before the outbreak.



Figure 27. Step 5, monitors the outcome of PRRS control program by tracking performance indicators such as time to baseline production.



*TTBP is the time in weeks, to reach the same number of weaned piglets as before the PRRS outbreak.



Normand et al., 2019,
ESPHM

5 Step for systemic PRRS control

Step 5. Implement and monitor preferred solutions

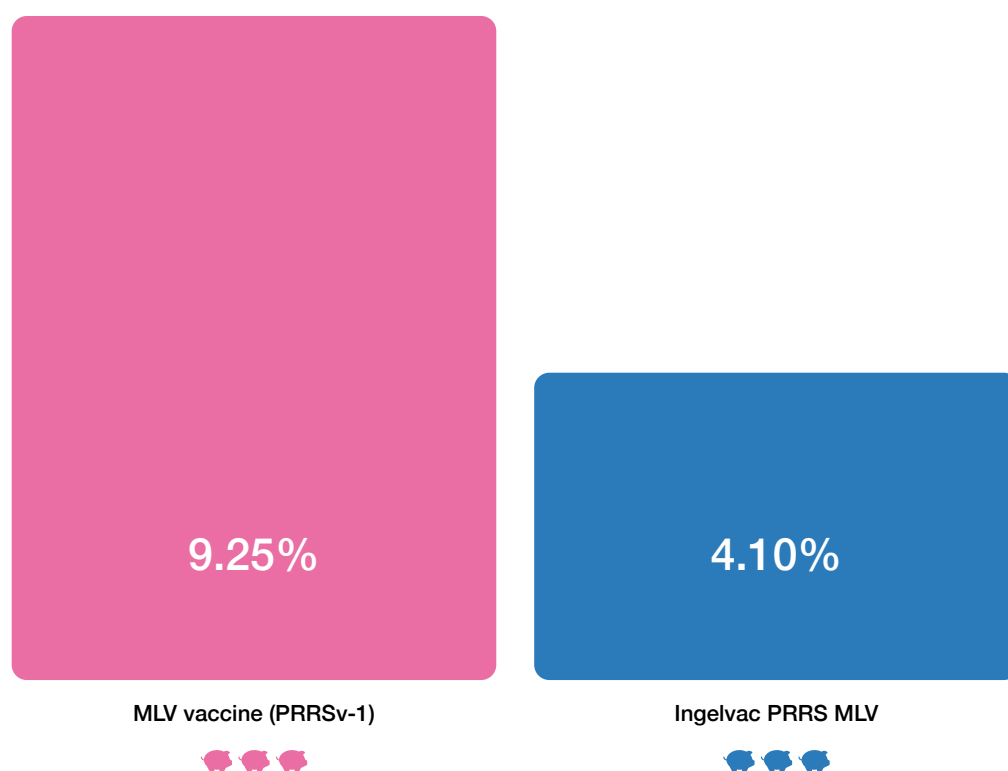
Mortality (Nursery)

Case report:

An important Objective for these endemically infected sow farms, located in Thailand, was to reduce pre-weaning mortality. Changing the piglet vaccination from a PRRSv-1 to Ingelvac PRRS MLV helped to reach the achieved target and lower mortality in nursery pigs.



Figure 28. Step 5, monitors the outcome of PRRS control program by tracking performance indicators such as nursery mortality.



Duangwhae *et al.*,
2017

5 Step for systemic PRRS control

Step 5. Implement and monitor preferred solutions

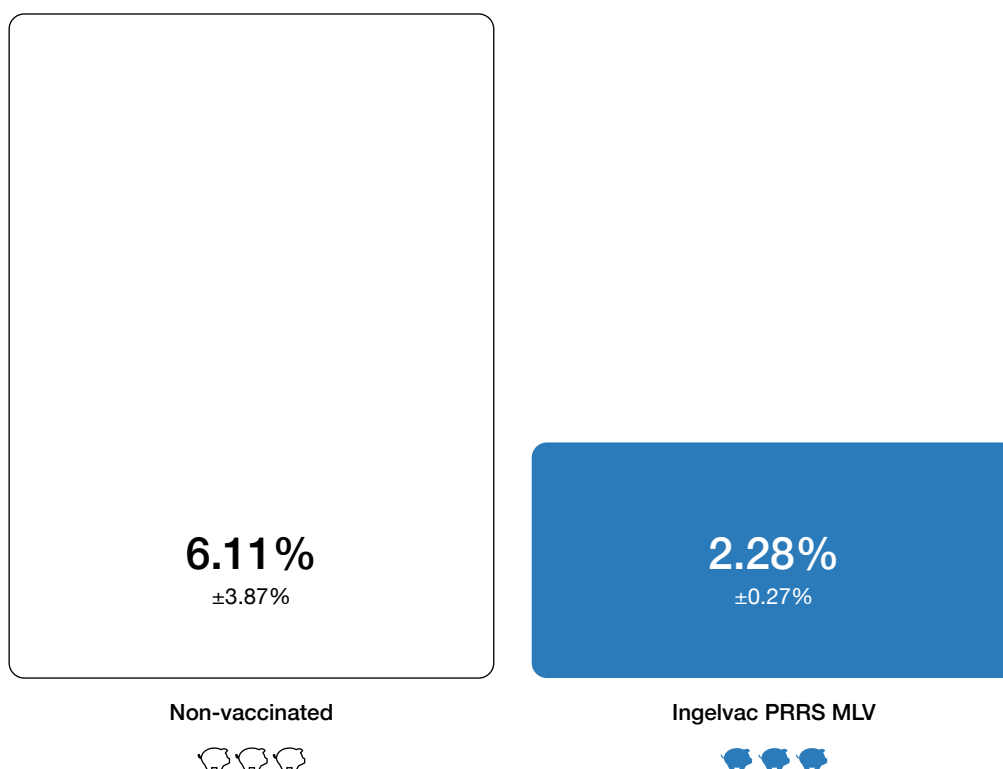
Mortality (Nursery)

Case report:

These data compile information on nursery mortality in 4 different farms, comparing nursery mortality with and without (before/after) piglet vaccination with Ingelvac PRRS MLV.



Figure 29. Mortality (Nursery).



Waddel *et al.*,
2008



Oropeza-
Muñoz *et al.*,
2012



Angulo *et al.*,
2012



Bae *et al.*
2017

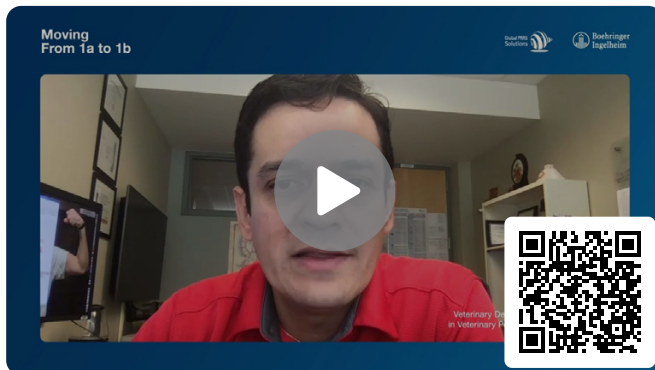
3.2 Moving up statuses: Moving up from 1A to 1B

Status 1A

Positive unstable at high prevalence:

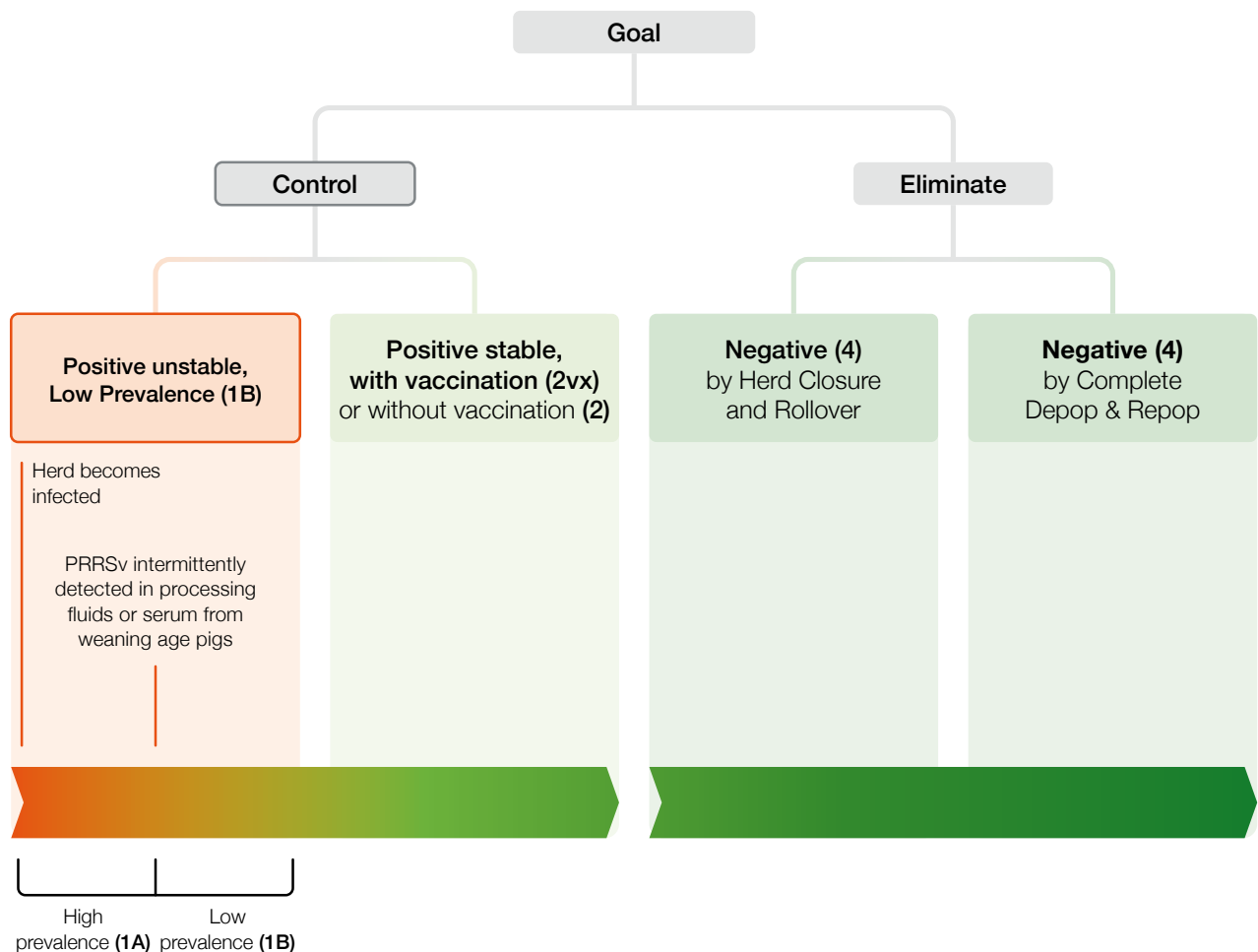
Typically shortly after an outbreak. This stage can last perpetually if no immune management and adjustments of pig flow are implemented. At this stage there is PRRSv circulation in abundance in the herd, and pigs

are clinically affected as evidenced by lower productivity. For example, there will be increased numbers of abortions, lower farrowing rate, weakly born piglets, lower survivability and decreased throughput (Number of pigs weaned per week).



G. Silva *et al.*, 2018

Goal: A herd that is not actively pursuing stability. The objective is to maintain category 1B with or without vaccination.



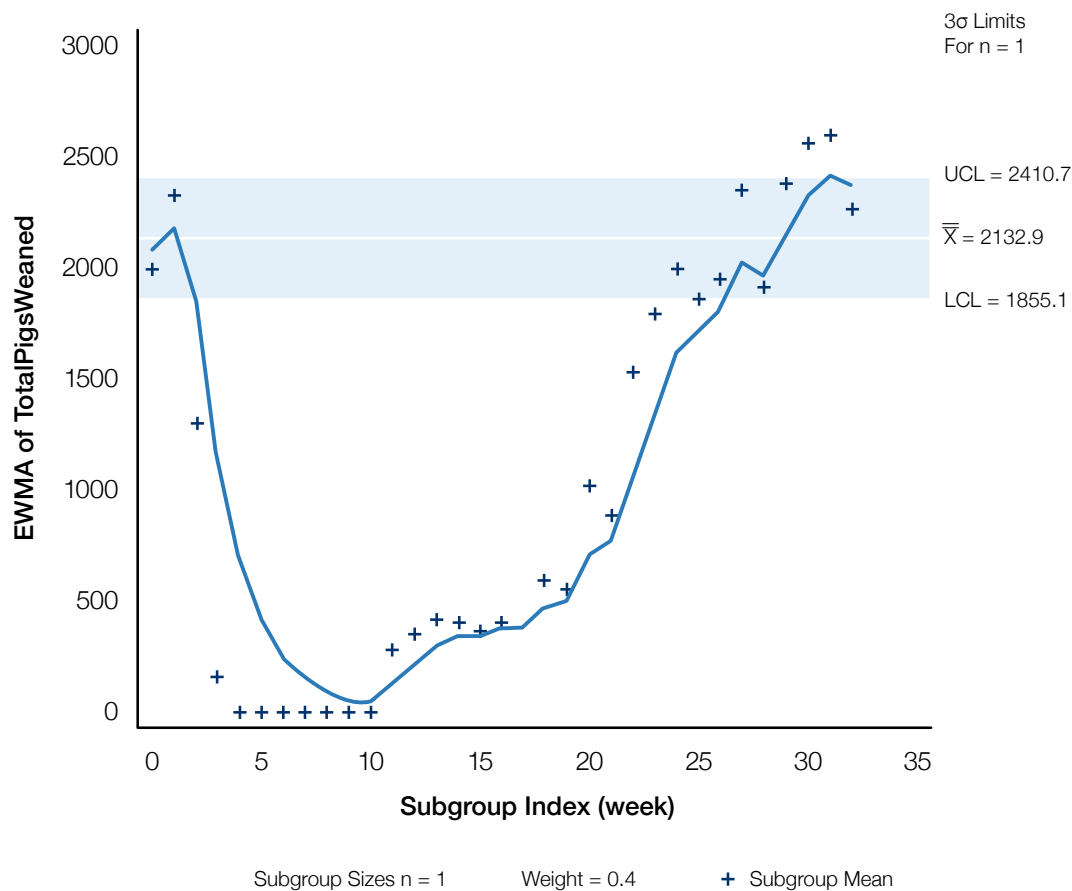
3.2 Moving up statuses: Moving up from 1A to 1B

Herds at **1A** should expect the following recovery time line. There should be opportunities to improve pig flow and immune management should this time line is not achieved:

- Recovery of abortion levels: 3-6 weeks.
- Recovery Stillbirths and Mummies: 12-16 weeks.
- Recovery number pigs weaned per week: 16-22 weeks.
- Start testing PCR-neg intermittently, raising CT values: possible 12-16 weeks, likely 26-30 weeks.
- Expect most PCR-neg suckling pigs: 30-36 weeks.
- Expect Negative-PCRs suckling pigs: 32-42 weeks.

There will be variation on clinical impact and time to recover productivity, depending upon herd immunity, pig flow, and management practices. The charts below demonstrate a breeding herd that had a severe production impact (left), and a breeding herd with mild productivity impact (right), measured as number of pigs weaned per week.

Figure 30. Exponentially weighted moving average chart for total pigs weaned.



Courtesy: Daniel Linhares

3.2 Moving up statuses: Moving up from 1A to 1B

Important to note that farms recover productivity BEFORE they quit shedding PRRSv. This is called “silent PRRSv” activity, also known as sub-clinical infection. If no serious effort and discipline are implemented, the virus will keep circulating, mutating, and evolving causing subsequent outbreaks in the breeding herd, and significant impact on growth performance downstream.

The key is to avoid contact between shedding and non-shedding (i.e., at risk) pigs, breaking the infection cycle. Key strategies to accomplish this and move from 1A to 1B include:

HERD IMMUNITY



Whole-herd expose the breeding herd, immunizing all females avoiding “pockets” of susceptible animals. The safest exposure method is a modified-live virus vaccine. Several controlled and field studies demonstrate the safety and efficacy features of PRRSv-1 MLV (ReproCyc PRRS EU/PRRS FLEX EU) or PRRSv-2 MLV (Ingelvac MLV) to achieve this goal.

When the goal is to control rather than eliminate PRRSv, many veterinarians choose to routinely vaccinate the breeding herd (i.e., every 3-4 months), keeping the herd immunity “high”. This strategy has been demonstrated to significantly reduce the clinical and productivity consequences of wild-type PRRSv introduction.

PIG FLOW



Load-close-expose (LCE) is a strategy to bring to the acutely infected herd as many gilts as possible, interrupting future introductions afterwards, and expose them to the same material used to expose the breeding herd (i.e., MLV vaccine). It is important that no gilts actively shedding PRRSv are introduced to the herd until there is evidence of consistent production of PRRSv-negative pigs at weaning. LCE is one of the most important and consistent strategies to achieve wild-type PRRSv control & elimination.

For herds that do not implement Herd Closure (i.e., continue to introduce gilts while still weaning PRRSv-positive pigs), it is crucial that gilts are immunized two (ideally three) months before being introduced to the breeding herd, assuring a high level of immunity. This ensures that when in eventual contact with the wild-type virus, the period of viremia and shedding will be transient.

BIO-MANAGEMENT AND BIO-CONTAINMENT PRACTICES

McRebel-like practices such as not holding pigs back for quality, changing needles/scalpels between litters, not cross-fostering pigs between litters, not stepping into the crates are important to make it harder for virus to transmit between pigs, crates, rooms, and week after week.

Find the 10 most important Biosecurity and management rules on the next page. The Swiss cheese concept emphasizes the importance of adding several Biosecurity layers in order to reduce the virus transmission.

Swiss cheese model

The Swiss cheese model consists of the 10 most important Biosecurity rules. By adding layer after layer, the virus transmission is effectively reduced.

Find more information for each rule including animated videos in the appendix or scan the QR code.



Swiss cheese model

Beside this general Biosecurity advice, online tools such as COMBAT allow more customized guidance.



1. Litter equalisation

2. Reduce cross-fostering

3. Farrowing pen management

4. Check needle management

5. Don't move sick piglets

6. Wean the whole room (all out)

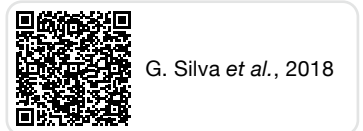
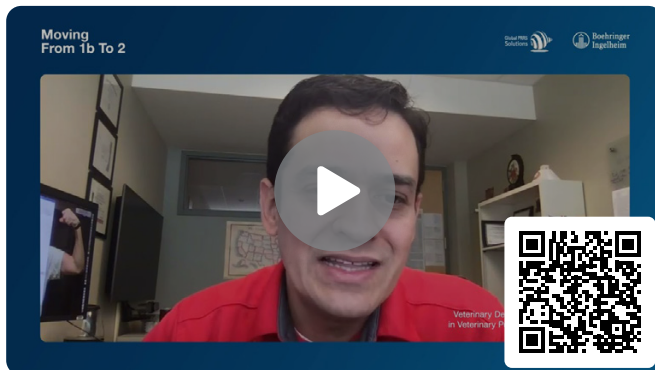
7. Strict batch production

8. No contact between age groups

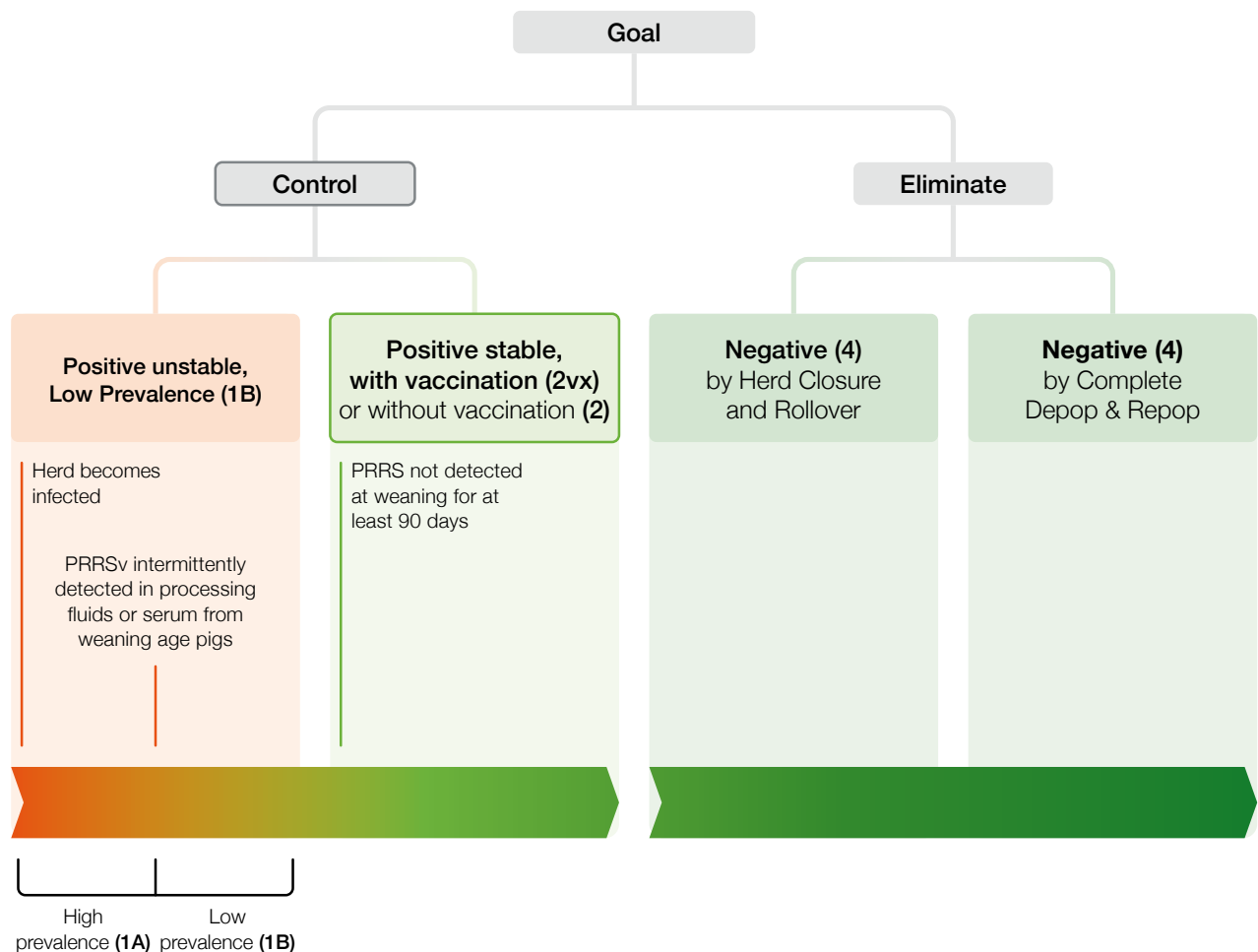
9. Avoid reinfection of sows

10. Check quarantine

3.2 Moving up statuses: Moving up from 1B to 2 (or 2vx)



Goal: A herd that is actively pursuing stability by means of vaccination.



3.2 Moving up statuses: Moving up from 1B to 2 (or 2vx)

Status 1B

Positive unstable at low prevalence: most likely the productivity and clinical aspect of the breeding herd will be “back to normal”. However, there is still virus “leaking” from the breeding herd at low prevalence. Most likely the production (and economic) impact of this low-level PRRSv activity will be more evident in the downstream flow, particularly after 8-10 weeks post weaning when levels of maternal immunity wane.

The strategies to move from **1B** to stable with (**status 2vx**) or without (**status 2**) vaccination are the same listed for moving from **1A**. The difference here is the intensity and duration of implementation. In other words, moving from **1A** to **1B** takes 8-16 weeks. Achieving stability, defined as consistently weaning PCR-negative pigs at weaning, takes an average of 33 weeks, ranging from 12 to 52 weeks. The large variability is due to PRRSv strains, herd immunity, pig flow, and management practices.



3.2 Moving up statuses: Moving up from 1B to 2 (or 2vx)

Key strategies to achieve stability are:

LONG HERD CLOSURE

Introduce non-shedding gilts when there is evidence of consistent production of PCR-negative pigs for a period of 13 weeks (3 months). Typically herd closure is associated with whole-herd exposure to a live virus, as described in the section **1A** to **1B**. MLV vaccine has been demonstrated to be an efficient exposure method, associated with lower productivity impact in the herd compared to using the resident live virus.

BIO-MANAGEMENT

At low prevalence (stage **1B**), there will be small clusters of PRRSv-positive pigs in some crates of some (not all) rooms. Thus, it is important to educate farm personnel to keep internal biosecurity high. An example of practices include avoiding transfer of pigs between rooms or even between crates. Another great practice is decontaminating hands, boots, coveralls when moving between rooms. To achieve the best PRRS control check and assess your Biosecurity and management with free online tools such as COMBAT.

MOSS

Because PRRSv circulation at this stage is sub-clinical, strategic monitoring of suckling pigs for PRRSv is a key aspect of the PRRSv management program. A combination of PF, serum, tongue tips, and FOF can be implemented as discussed in section 1.0 (page 7).

PIGLET MOVEMENT

Across rooms, or introduction of naïve or shedding gilts into the breeding herd when there is low level of wild-type circulation in the breeding herd can lead to PRRS outbreaks.

The difference between status 2 and 2vx is the ongoing use of MLV vaccination. Herds on 2vx will have ongoing MLV exposure in gilts or the breeding herd.

Herds vaccinating gilts should do so 2-3 months prior to entering them in the breeding herd, allowing proper time to establish a protective immune response.

Herds vaccinating sows typically do so 3-4 times per year. It is expected that the PRRSv-stable breeding herds produce viremic piglets due to MLV for 1-2 weeks post sow vaccination.

Sow herd stabilization and improvement of Farm performance with whole herd vaccination.

3.2 Moving up statuses: Moving up from 1B to 2 (or 2vx)

A case report



Case history

This Malaysian PRRS endemic farrow-to-finished farm (250 sows) has been using Ingelvac® PRRS MLV only in sows for half a year and was able to achieve a stable PRRS status by weaning PRRS negative piglets. No PRRS vaccine was given in piglets. Recently, PRRSv-1 and PRRSv-2 was found in 4 weeks old and sick piglets. Ingelvac® PRRS MLV piglet vaccination was implemented in 2 weeks old piglets over a period of 6 months.

Monthly mortality percentage for due to wean piglets, nursery- (1-2month old) and fattening pigs (>2 months old) were collected. Serum of 4 weeks old piglet were tested for PRRSv by PCR.

Results

After starting Ingelvac® PRRS MLV vaccination in piglets, an improvement in reduction of mortality at all stages was documented. The mortality was reduced by 7.4%, 19.6% and 57.3% in piglet, nursery, and finishing pigs (>2 months old) respectively (Table 11). With a whole herd vaccination protocol, the overall mortality decreased from 6.73% to 3.74%, that accounted to approximately 44% improvement.

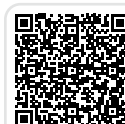
Other than that, pooled serum sample for 4 weeks old pigs were negative compared to before piglet vaccination. Implementing a whole herd vaccination protocol (vaccinating sows and piglets) resulted in significant reduction of mortality.

Summary

In this field trial, whole herd PRRS vaccination with Ingelvac® PRRS MLV was able to reduce mortality in farm due to PRRSv infection. The largest improvement was seen in fattening pigs (> 2 months) where mortality was significantly reduced by over 50%. After implementation of whole herd vaccination, the farm became PRRS stable again by weaning PRRS negative piglets.

Table 10. Mortality (%) in growing pigs.

Mortality (%)	PRRS Vaccination (only Sows)	PRRS vaccination (sows+piglets)	Diff.%	p- value*
Piglet	10.14	9.39	-7.4	0.405
Nursery	10.51	8.46	-19.6	0.405
Finisher	3.34	1.43	-57.3	0.014
Total	6.73	3.74	-44.3	0.033



Shen Rong Tai *et al.*,
IPVS 2022



3.2 Moving up statuses: Moving up from 2vx to 3 and 4

Positive stable: no clinical signs, no evidence of wild-type circulation in the herd (**2vx** herds allow some MLV virus detection following the breeding herd vaccination).

When the goal for the herd is to achieve Control, **status 2** or **2vx** is the final destination.

When the goal is to Eliminate the virus without depopulation-re-population, the final destination is **status 4**, which is obtained after **status 3**.

This move (from 2/2vx to 3 - 4) should be done if and only if:

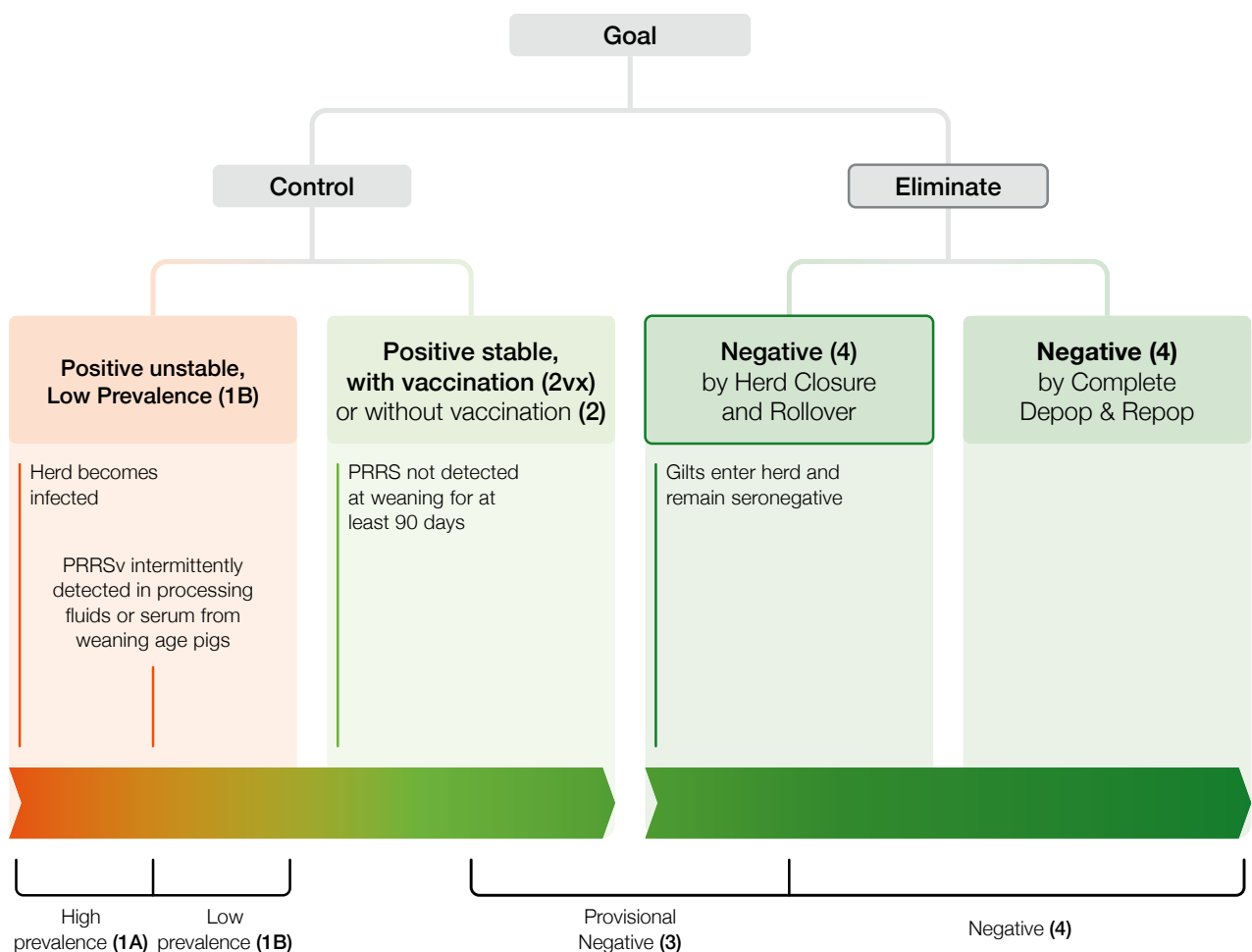
- There is certainty of no shedding (zero prevalence), which requires proper diagnostic testing as described in section 2.0 (page 55).

- There is low risk of re-introduction of new PRRSv strains, with expectation of maintaining the herd free of outbreaks for at least 2-3 years. This requires a solid Biosecurity plan (e.g. COMBAT) and understanding of biosecurity hazards in place.

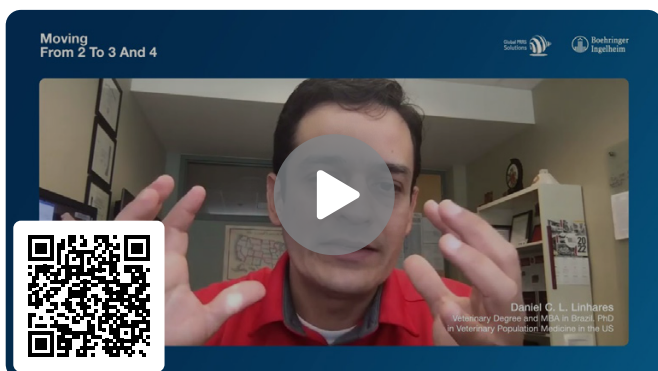
Moving to **status 3** requires introduction of naïve gilts to the non-shedding breeding herd.

Status 4 (naïve herd) is achieved when there is no serologically positive sow in the herd, which is achieved with time — typically ~2 years considering a sow replacement rate of ~50%.

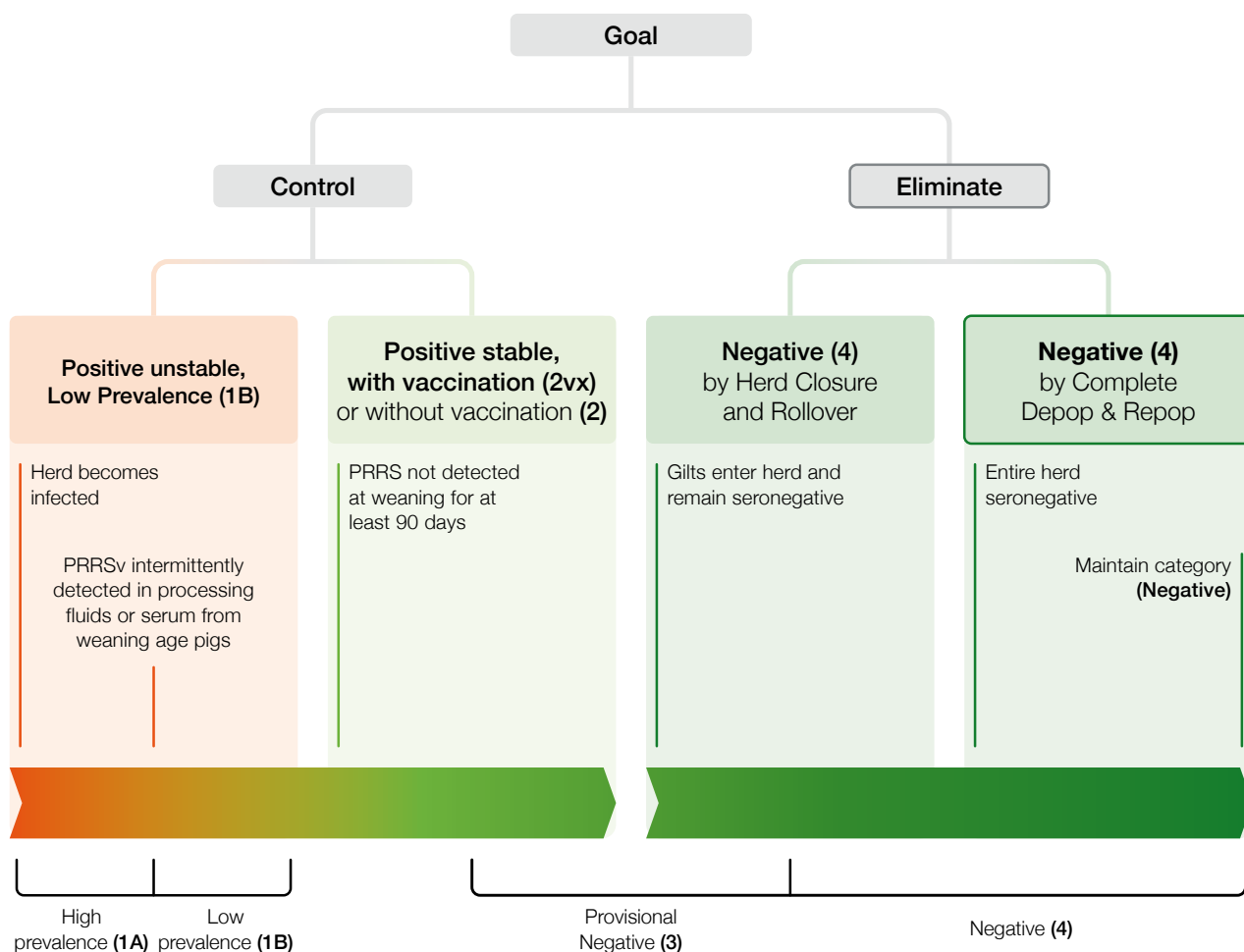
Goal: A herd that is actively pursuing a negative PRRSv status by herd closure and rollover.



3.2 Moving up statuses: Moving up from 2vx to 3 and 4



Goal: A herd that is actively pursuing a negative PRRSv status by complete depopulation & repopulation.



3.2 Moving up statuses: Moving up from 1a to 2vx and then to 3 and 4

A case report



Use of tongue tip fluids to monitor PRRS prevalence during an eradication in a Spanish farrow to feeder farm.

Case history

A 17-year PRRSv negative farrow-to-feeder farm producing 18-kg pigs, became infected with PRRSv-1. The outbreak negatively affected performance parameters such as pigs being born alive and pigs weaned. Disease losses in the farm were mainly located in nursery, with mortality of up to 20%. Clinical signs were compatible with PRRSv infection. Furthermore, other clinical signs were present in the nursery indicating secondary or concomitant infections such as meningitis and diarrhoea. Necropsy confirmed presence of bacterial coinfections such as *Glaeserella parasuis*, *Pasteurella multocida*, *Actinobacillus pleuroneumoniae* and *Bordetella bronchiseptica*. As part of the 5 Step process to systemically control the disease, PRRSv-1 eradication was identified as the main goal (Step 1). 18 months after the outbreak, an eradication plan was put into place.



Example picture of PRRS-related dead born piglets in an acutely affected Sow farm. Courtesy E. Marco

3.2 Moving up statuses: Moving up from 1a to 2vx and then to 3 and 4

A case report

The eradication plan protocol

Systemic monitoring of stillborn piglets using tongue tip fluids was implemented. Samples were found initially PRRSv positive by PCR suggesting vertical transmission. All sows were vaccinated with a PRRS MLV vaccine and Ingelvac CircoFLEX and revaccinated 4 weeks later. Whole herd PRRS vaccination was implemented to reinforce immunity and to stop any vertical transmission from sows to piglets or horizontal transmission between sows. To keep all sows on the same immune status, mass vaccination was repeated every four months.

Nurseries and gilt development unit (GDU) were emptied, thoroughly cleaned and disinfected. The use of the smallest nursery buildings (less than 50 meter from the breeding herd facility) was changed to avoid the presence of weaned pigs close to the breeding herd. The main nursery buildings (230 m away from the breeding herd) were emptied in order to stop PRRSv recirculation and to reduce infectious pressure of all concomitant pathogens (viral and bacterial). Emptying of this building was repeated a second time, since PRRSv positive piglets were still being weaned. It was kept empty until the flow of piglets was completely PRRSv negative.

All gilts in the on-site GDU were introduced into the breeding herd and bred regularly. Once cleaned and disinfected, the on-site GDU was loaded with PRRSv negative gilts for a 6-week period. Gilts were bred in the same GDU. The external GDU was also loaded with gilts for an extra 10-week period to be bred in the same building.

The three stages of production, breeding, nursery and off-site GDU are now being managed as three independent farms and biosecurity measures are applied to avoid any cross-contamination. Personnel are exclusive to each stage.

In case that more than one stage needs to be visited (e.g., veterinarians, feed or material deliveries), PRRSv negative stages are visited first, before any contact with PRRSv positive units.

The farm will be closed for 200 days. After that period, pregnant gilts located in the external GDU will be transferred to the breeding herd at the time of farrowing. The on-site GDU will be filled with 18 kg PRRSv negative animals.



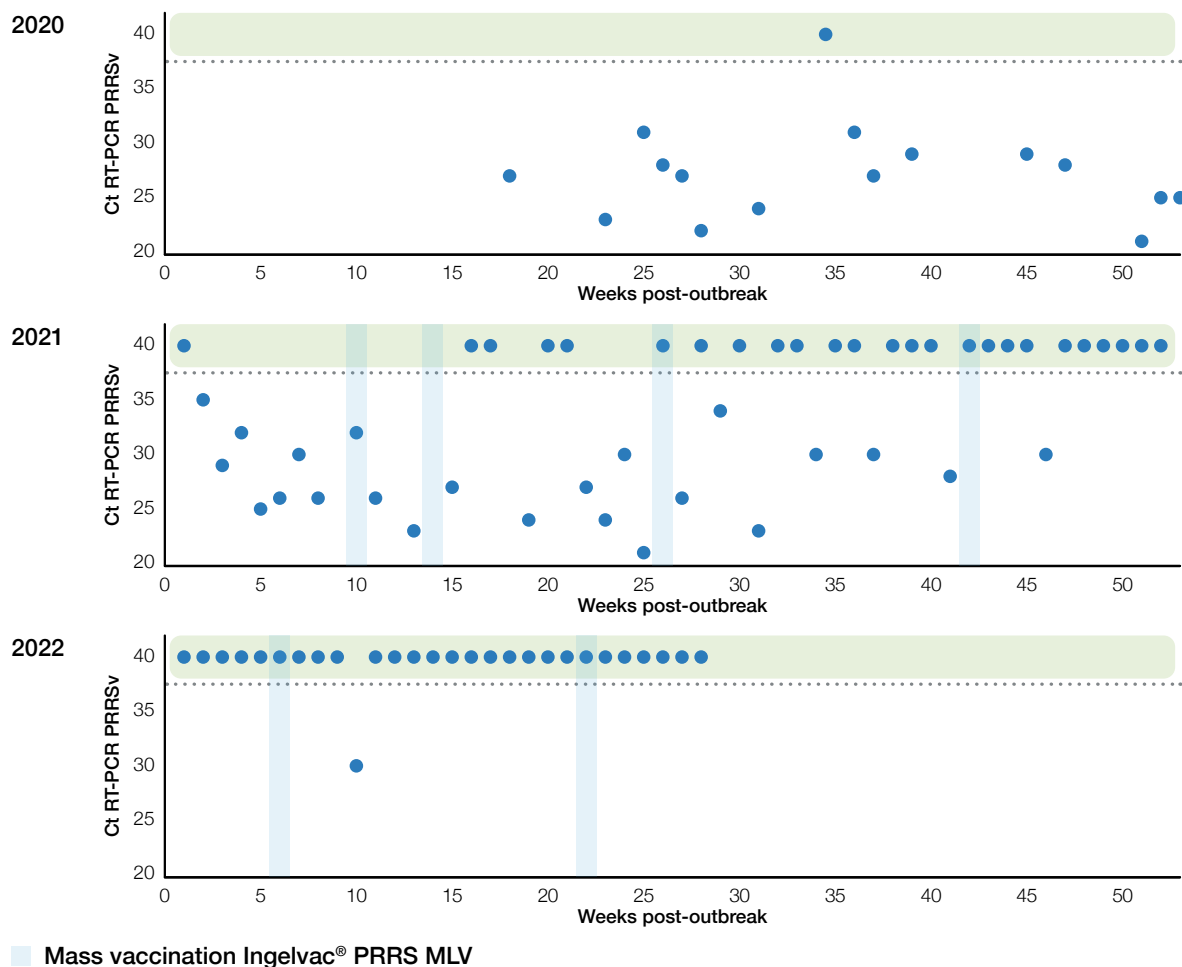
3.2 Moving up statuses: Moving up from 1a to 2vx and then to 3 and 4

A case report

Current status

- The farm will start breeding PRRSv negative gilts from the external GDU.
- Tongue tip fluids have been PRRSv negative by PCR for 14 weeks.
- Serum from weaned pigs (n=30) were PRRSv negative by PCR. Sample size will be increased to detect lower prevalence.
- Animals being weaned to external nurseries are PRRSv negative by PCR at serum 3-4 weeks after weaning. However, pigs weaned in the farm main nursery were found viremic 3-4 weeks after weaning with high levels of viremia towards the end of the nursery period. Due to this information, it was decided to empty, clean, and disinfect the nursery a third time to avoid any potential risk of rebreaks.
- As today, the number of live born and weaned piglets have increased since the start of the plan. Fertility has stabilized and the mean mortality in the nursery was decreased to 4.87%.

Figure 31. CT RT PCR PRRSv Vs. Calendar week.



Summary

PRRSv infection causes economic losses, regardless of strain or prior immunity in pig populations. Thus, it is imperative to implement practices to decrease the PRRSv activity in affected swine populations, allowing pigs to reach their full productivity potential. Depending on the scenario and business structure, it makes sense to target complete virus elimination from some herds, keeping the pig populations naive and avoid any PRRSv impact. PRRSv elimination plans hinge upon building strong herd immunity, oftentimes using MLV vaccination associated with changes in pig flow, including herd closure and implementation of strict bio-management practices.

A key component of the PRRSv elimination plan is diagnostic monitoring using tools extensively discussed in section 1.0 (*page 7*).

However, in some situations, it is nearly impossible to keep herds negative; thus, some veterinarians recommend keeping the herd immunity high, lowering wild-type PRRSv activity and impact. In such cases, the goal is to reduce the viral diversity (i.e., the number of circulating strains) and lower the prevalence of PRRSv in all age groups. This requires constant immunization of the breeding stock and growing pigs, and attention to pig flow, avoiding mixing pigs of different PRRSv statuses. Regardless of the strategy (control versus PRRSv elimination), the 5-step process is a great tool to aid veterinarians in fine-tuning their plan by identifying desired goals; determining the current PRRSv status; understanding current constraints; developing solution plans; and implementing such plans while monitoring the outcomes.

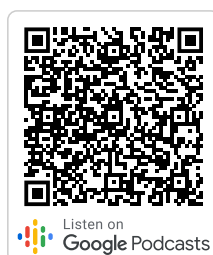
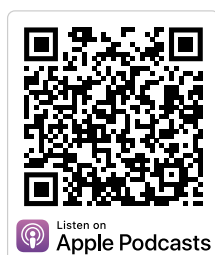
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Frequently Asked Questions

Diagnostics

When classifying farms for PRRS, can we use S/P ratios to define the level of protection?

It is true that S/P ratio can be related to the amount of IgG Antibodies measured in the ELISA test. It is important to understand, that these antibodies are not related to protection and therefore can not define a level of protection.

ELISA results can only be used to determine if a previously naïve animal was in contact with PRRSv (vaccine or field virus) and not to determine the degree of immunity, neither in an animal nor in a herd. The only immune parameters correlated to protection are neutralizing antibodies and cell mediated immunity (measured by Interferon γ) as described in (Figure 34).

Currently there are no commercial tests available to measure the amount of these protective neutralizing antibodies and immune cells. Make sure to use a vaccine that induces a sufficient amount of cell mediated immunity such as Ingelvac PRRS MLV, that helps to effectively protect against the negative effect PRRS.



Shen Rong Tai et al.,
IPVS 2022

Cellular immunity and PRRS control in growing pigs.

Regardless of the challenge virus, vaccination of pigs effectively reduced the level of viremia, the lung lesions, and of the PRRS antigen within the lung lesions. The induction of virus-specific **interferon- γ secreting cells** by the PRRS vaccine produced a **effective immune response**, leading to the reduction of PRRSv viremia (serum viral load).

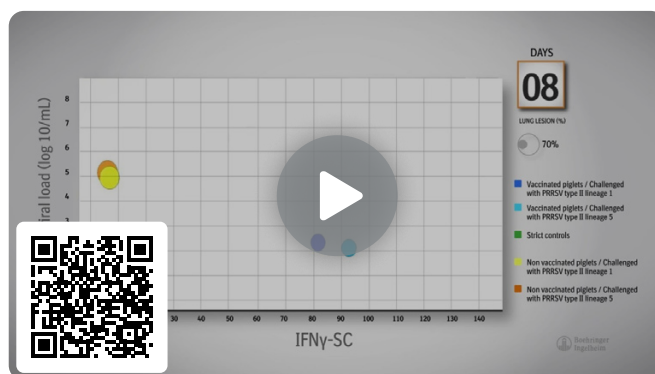
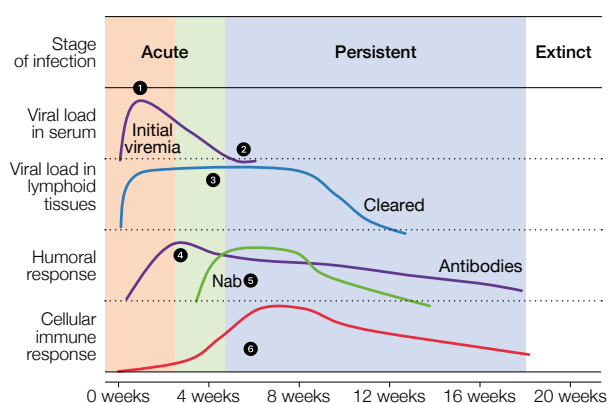


Figure 32. General immune response to PRRSv.



① PRRSv replicates in lung Macrophages resulting in **viremia** by 6-12 hours post infection and may last for several weeks **despite presence of Antibodies**.

② Later during the infection, virus replication subsides and can no longer be detected in blood and lungs.

③ At this stage, PRRSv replicates in lymphatic tissues. Replication slowly decays until the virus becomes extinct. This may last longer than **250 days** post infection.

④ The **initial Antibodies** can be detected 7-9 days post infection. They are quick but **not protective**.

⑤ **Neutralizing Antibodies** (Nab) appear later at day 28 after infection but are important for **protection**.

⑥ **Cellular immunity** is key for an **effective protection**. This response takes at least two weeks and is initially low. But once the cellular immune response took off, **the viral load in tissue drops** and gets cleared eventually.

How many tongues do I need to extract enough fluids for sound diagnostics?

For older animals (>7 days of age), you might need more than 20 tongues in a bag. Current study results suggest, if samples are frozen (-20°C) and thawed, 14 to 45% more liquid can be extracted. These results also showed that Ct values didn't change after adding 1mL PBS into a bag of 30 tongue tips.

What is the best way to early detect PRRSv infection in a breeding herd?

It only takes about 24h for PRRSv to replicate and become viremic. However, depending on herd immunity and on the virus strain, transmission can be slow (like in *Mycoplasma hyopneumoniae*) before the outbreak fully blows out. Thus, in an ideal world people should test various sample types daily. However, this would be time and cost prohibitive.

One efficient way to monitor the breeding herd for PRRSv activity is watching closely clinical and productivity outcomes such as keeping track of sows off-feed, number of abortions, neonatal pig losses (stillbirths and mummies), and pre-weaning liveability. Whenever there is any spike in these metrics, diagnostics should be done to confirm or rule out PRRSv infection. Practical and effective options include PCR testing of processing fluids, tongue tips, and aborted fetuses.

Diagnostics

Using SoundTalks to monitor GDU for signals of respiratory disease can also allow for early detection of PRRSV introductions.

Tongue tips seems to be a nice emerging sample type. When should I use it, and when to stick with processing fluids for PRRSV detection?

Processing fluids are perhaps the most practical and sensitive method to screen herds (practicing castration) for PRRSV circulation. Tongue tip fluids are a great alternative for herds not castrating. It is also a great sampling approach when targeting tongues from stillbirth pigs when the intent is to assess vertical transmission.

I am considering pooling my family oral fluid samples (FOF) but I'm afraid of the potential dilution effect. I can only afford 5 PCR tests. Is it better to test 5 individual samples, or collect more and pool (e.g., 5 pools of 25 samples)?

Mathematical modelling of FOF samples pooled in different scenarios, and validation with field samples demonstrate that you are much more likely to detect PRRSV in pooled samples than in individual samples when you are pooling to increase your coverage (i.e., number of crates sampled).

Assuming a prevalence of about 8% in the room (5 PRRSV-positive crates out of 56), your probability of detection FOF with 5 samples is only 36%, as compared to 93% when you do 5 pools of 5 (25 total samples) in the same room and condition.

Control

In case my sow farm breaks with PRRS, what are the 3 most important tasks?

- Check sow herd stabilization and vertical transmission.
- Apply whole herd vaccination.
- Check and improve internal and external biosecurity.

Check sow herd stabilization and vertical transmission

The epidemiologic cycle of PRRS in the farm is usually perpetuated by the existence of vertical transmission events. In other words, as far as viremic piglets are born, the chances for controlling of PRRSV infections downstream in the production cycle (nurseries, fattening units) are seriously diminished. Therefore, stabilisation of sows is a crucial element to reduce the number of infections in the farm.

To achieve this objective, the key is starting with a good planning of the replacement policies. Performing a good biosecurity program for gilts with adequate quarantine facilities and testing of animals to avoid entering infected individuals is a first step. Then, gilts should be acclimated by vaccination with a MLV to ensure that they have immunity against the virus before the first insemination (two doses recommended). Once gilts are entered in the breeding stock, immunity must be maintained by means of recall vaccinations every 3-4 months.

Internal biosecurity measures are needed to avoid recirculation of the virus between different production phases. Moving animals against the production flow, sharing personnel or materials between different production phases or not changing needles when vaccinating sows may contribute to the spread of the virus. Testing of suckling piglets by RT-PCR and sequencing will help to monitor the evolution of the program (see section 1.0, page 7) to understand how PF, FOF or TTF help to understand PRRSV circulation).

In summary, internal biosecurity aims to make it hard for the virus to move between crates, rooms, barns, and production stages. Practical examples include implementing bio-management practices such as hygiene, changing coveralls, washing hands or using gloves when moving between rooms.

Remember the basic rules:

- Do not enter infected gilts, acclimate them.
- Implement a recall vaccination program.
- Minimize PRRSV transmission from sows to piglets.

Apply whole herd vaccination (Sows and piglets)

The goal of your PRRS control program is to reduce the number of strains and the amount of wild type virus in your herd. The whole herd vaccination plan reduces the circulation of PRRSV.

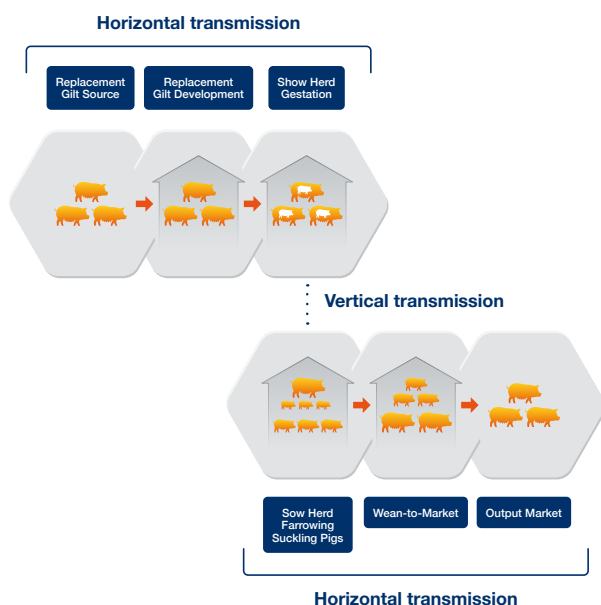
We recommend to vaccinate sows 3-4 times per year by mass vaccination. That helps to keep all sows at similar immune status (check if you combine vaccines by mixing PRRS MLV with other compatible vaccines).

During quarantine, vaccinate your gilts twice, 30 days apart.

Control

Vaccinate piglets at 3 weeks of age. In this age you run less risks of maternal interference of vaccinated sows. Mixing vaccines help to reduce injections and labour.

Figure 33. Infection chain.



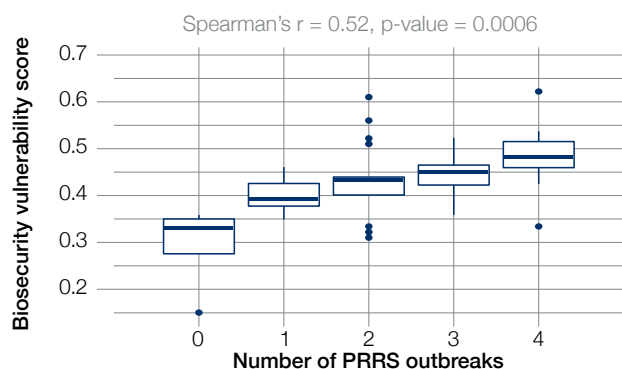
- Additional information, Meet the Expert:



Check and improve internal and external biosecurity

Beside vaccination, Biosecurity is the other key pillar for effective PRRS control. It is common sense, that effective Biosecurity standards help to reduce PRRSv circulation and can prevent PRRSv breaks.

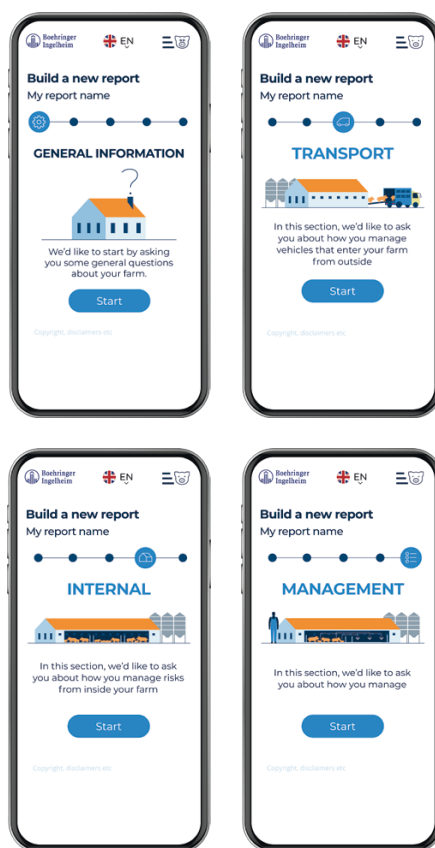
Figure 34. Biosecurity standards & PRRS outbreaks.



Reducing the risks of PRRS transmission (both within the farm and from outside introduction) can be a challenge, but what are the things I can do?

General biosecurity guidelines are in public domain.

Nowadays free online tools help to check and improve your Biosecurity status with customized advice. They further allow a monitoring and comparison of biosecurity statuses separated into main categories (general, external, transportation, internal management) reflecting the latest scientific knowledge.



Visit COMBAT

Control

What are specific key factors associated with recovery of breeding herds from PRRSv outbreaks?

Based on epidemiological studies, factors associated with shorter time-to-stability and lower production impact are:

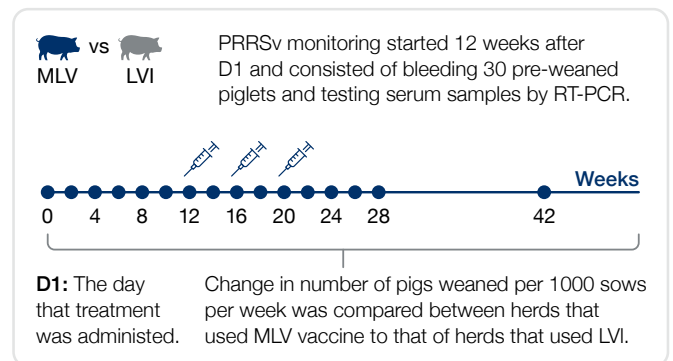
- Prior immunity, whether it is MLV-derived or from a previous outbreak. Naïve herds take longer to recover and have higher losses.
- Herd closure increases the success rate to achieve stability.
- Deliberate whole-herd exposure to any type of replicating virus (wild-type or MLV) is better than not exposing the herd. MLV use is associated with less severe production impact from the wild-type infection, while exposing to the live virus is associated with quicker time-to-stability.
- Batch farrowing system was associated with better productivity.
- Timing bio-management practices: sooner = better.
- PRRSv genotype: infection with novel strains to the herd, or to multiple strains at once is associated with worse outcomes.

To achieve a fast return to baseline production (TTBP) after a PRRS break should I use live virus inoculation (LVI) or vaccines (MLV)?

To control and eliminate PRRSv from breeding herds, some veterinarians adopt load-close-expose, which consists of interrupting replacement pig introduction for several months and exposing the pigs to a replicating PRRSv. This was a prospective field investigation that followed 61 breeding herds acutely infected with PRRSv that adopted one of two exposure programs: modified-live virus (MLV) vaccine or live-resident virus inoculation (LVI).

Treatment groups (load-close-expose with MLV or LVI) were compared for:

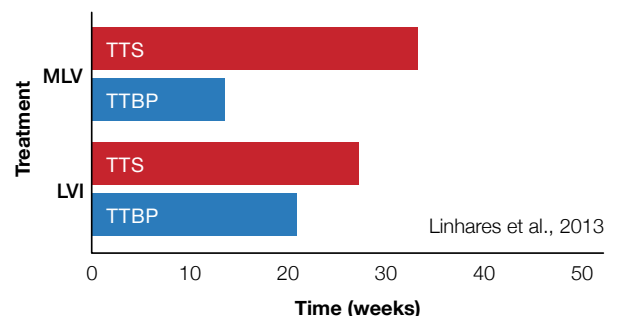
- **Time-to-PRRSv stability (TTS)**, defined as time in weeks needed to produce PRRSv negative pigs at weaning.
- **Time-to-baseline production (TTBP)**, defined as time to recover to the number of pigs weaned per week that herds had prior to PRRSv outbreak.



Herds were assumed to achieve “TTS status” when there was a failure to detect PRRSv RNA in serum of pre-weaning pigs by RT-PCR tested monthly over a 90-day period.

The median TTS among participating herds was 26.6 weeks (25th to 75th percentile, 21.6-33.0 weeks). The overall TTBP was 16.5 weeks (range 0-29 weeks). The magnitude of production losses following whole-herd exposure averaged 2217 pigs not weaned/1000 sows and was correlated with TTBP.

Figure 35. PRRSv Load-Close-Homogenize programs.



Herds in the MLV group recovered production sooner and had less total loss than herds in the LVI group. TTBP and TTS were significantly shorter and the total loss was significantly less in herds assisted by a specific veterinary clinic and herds that were infected with PRRSv in the 3 years prior to the study. This study provided new metrics to assist veterinarians to decide between methods of exposure to control and eliminate PRRSv from breeding herds.

Control

Pattern of PRRSv RNA by RT-PCR detection	LVI	MLV
Pigs not weaned/1000 sows from PRRSv detection to exposure (mean \pm std. error)	678.4 \pm 106.0	335.3 \pm 141.4
Pigs not weaned/1000 sows following whole-herd exposure (mean \pm std. error)	2665.0 \pm 313.0	1222.2 \pm 395.3
TTBP (median and 25th to 75th percentile)	21 (13, 24)	10 (0, 15)

Is using a “homologous” LVI instead of MLV vaccine in breeding herds to achieve time to negative pig production less expensive to use?

No. The MLV protocol is economically advantageous compared to the LVI protocol in the ‘LCE’ methodology for breeding herd stabilization. The cost to expose animals is greater with MLV stabilization programs; however, the total opportunity cost per 1000 sows for the LVI program was approximately \$26,000 more costly vs. the MLV program. The higher opportunity cost of the LVI program is due to the eight-week longer time period to achieve baseline production compared to the MLV program.

Vaccination

Are LVI’d replacement gilts and/or sows better protected against a challenge with a wild-type virus of similar “type/classification” than a challenge with a wild-type virus of a different “type/classification?”

No. Data would suggest that LVI offers partial protection against homologous and/or heterologous challenge. Of note is the difficulty to manage LVI in the field without developing endemic perpetuation of wild-type PRRSv circulation within populations and that true homology in the field is a fleeting occurrence.

Does heterologous vaccination provide good cross-protection with contemporary, highly virulent strains?

Yes. There is no correlation between strain-vaccine heterology and level of protection.

The vaccination does reduce the negative impact of any wild-type strain circulating in the farm. Any vaccination alone will not bring mortality from 20% to 1%, but it can reduce at least some 5 to 10 percentage points in the mortality, which is a truck-load of money.

When doing a herd closure & mass exposure, is it safe to expose all sows to modified live vaccines (MLV), or should I stage vaccinations according to breeding age groups?

It has been shown that reproductive losses associated with MLV exposure are transient and of small magnitude. Exposing all breeding sows to Ingelvac MLV shortly after an outbreak with wild-type is proven to significantly reduce the clinical consequences of PRRSv, as compared to relying on natural transmission of the wild-type Moura *et al.*, 2019.

I am weaning pigs from stable sow farms, and they are placed in a moderately dense region. Pigs look fine, but they are typically ELISA-positive at the end of finishing. Should I consider vaccinating them?

Vaccination of pigs around weaning with Ingelvac MLV has been proven in several studies to significantly reduce the clinical consequences of wild-type PRRSv natural exposure (i.e., improved ADG, liveability). In addition, MLV vaccines are a great bio-containment tool that significantly reduces the duration, magnitude, and viral diversity of wild-type shedding.

Therefore, vaccinating piglets with MLV is a great strategy to treat and prevent PRRSv in grow-finish, while also reducing the regional pressure of infection of wild-type PRRSv, which, in turn, decreases the probability of outbreaks in nearby sow farms.

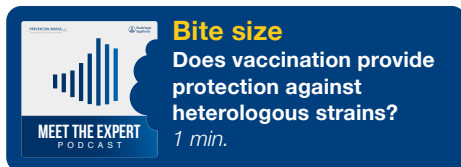
What are the advantages of bringing the herd back to Negative following an outbreak, as opposed to keeping the herd immunity active by keeping it on the Stable category?

Healthier pigs require less labour, less antibiotics, and are more efficient growing. Keeping herds in the Stable category is a strategy adopted by some production systems facing several outbreaks (3+ in a 5-year span). However, when the level of biosecurity is good enough to push outbreak incidence to one outbreak per 3+ years, it is economically advantageous to go Negative. This leads to a cleaner downstream flow, reduces the regional pressure of infection, and reduces the chances of re-exposure of the breeding herd through indirect contact with the grow-finish herd. Also, having PRRSv-negative flows helps to improve the culture of ‘biosecurity matters, prevention works, healthy pigs’ in the system, which can also be applied to Mycoplasma, Influenza, Coronaviruses, and other pathogens.

Vaccination

Why is vaccination of sows not enough to provide protection to growing pigs?

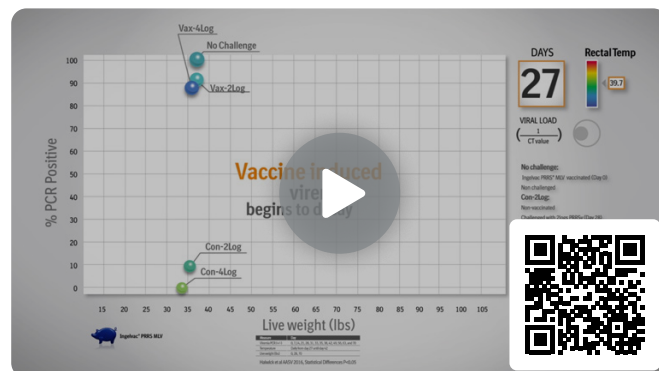
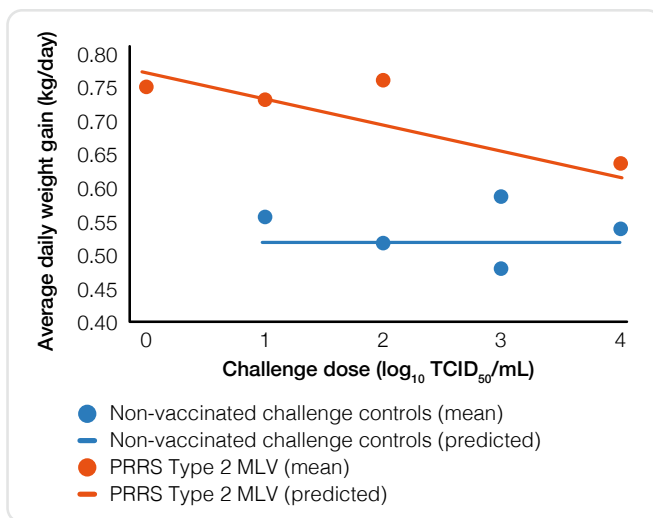
Sow vaccination is not enough to protect piglets. Similar to *Mycoplasma hyopneumoniae* and PCV2, sow vaccination is a great tool to build herd immunity in the breeding herd. However, the maternal immunity wanes rapidly within a few weeks after weaning, and piglet vaccination is essential to build active, long-lasting immunity in grow pig herds.



Why is it important to reduce the PRRS viral load in my farm?

Not only the PRRSv strain but also its quantity is correlated to clinical problems. Beside choosing an efficacious vaccine and making sure all animals are vaccinated properly, biosecurity and management practices help to reduce the spreading.

Haiwick *et al.*, 2018, demonstrated a measurable negative impact on ADWG in the non vaccinated and challenged groups with no difference across all challenge doses showing that already small amounts of virus has a negative impact. As compared to the non vaccinated challenged controls (blue dots), there was a significant increase in ADWG ($P < 0.05$) of vaccinates (orange dots) in the 3, 2 and 1log groups, and at $P < 0.07$ in the 4log group.



Can a properly vaccinated breeding herd have clinical PRRS breaks when challenged with a new, non-resident wild-type virus?

Yes. Cross-protection or heterologous protection exists, but is not complete or 100%. While vaccine-derived immunity can mitigate the consequences of infection and reduce clinical disease, infection is not prevented and horizontal and vertical transmission of virus can still occur. Transplacental transmission of virus in the immune pregnant female is also not completely prevented. Infection of in-utero fetuses prior to birth can occur. It is important to note that homologous protection does not offer complete protection or sterilizing immunity. Studies demonstrate that protection from homologous exposure is not complete and animals can become re-infected and generate viremia. Moreover, recent field data (Trevisan *et al.*, 2022) demonstrates that co-infection with multiple PRRSv strains is common in the field. As PRRSv continues to mutate and recombine, it is expected that there will be a cloud of different viruses co-circulating in the herd, making the homologous concept unlikely.

Are clinical PRRS breaks less severe in a properly vaccinated breeding herd when challenged with a new, non-resident wild-type virus?

Yes. In breeding herds that are properly vaccinated and have a level of herd immunity against PRRSv the clinical consequences of a new heterologous challenge are mitigated and the herd returns to baseline production performance quicker than herds that are not vaccinated.



Haiwick *et al.*, 2018

Vaccination

Is there a meaningful difference between a naïve sow farm and a properly vaccinated sow farm that gets infected with a new, non-resident wild-type virus?

Yes. Data demonstrates the immune status of breeding herds prior to an outbreak impact the consequences of a PRRS outbreak. Prior exposure/vaccination and development of uniform population immunity offers protection against the detrimental productivity impact of outbreaks. Vaccine derived immunity mitigates the consequences of infection and reduce clinical disease. PRRS outbreaks in breeding herds that are PRRSv-free immediately prior to an outbreak are more severe; as measured by born alive, pre-weaning mortality and take longer to return to base-line performance than herds that have a level of population-based immunity at the time the break occurred. These data indicate it is economically beneficial to maintain a level of population immunity for PRRS in breeding herds at risk to PRRSv infection.

Is there a value of routine pig vaccination in hog-dense, high-PRRS-prevalence areas in relation to reduced signs of clinical disease and improved performance?

Yes. Several field research studies demonstrate economic and biologic impact of vaccination for the control of PRRSv in growing pigs with improved health and performance as measured by improved average daily gain, reduction in mortality and culls and improved percent of animals reaching prime market targets. Even with the variables of field research studies there are numerous examples of the economic and biologic benefit of vaccination for the control of PRRSv in growing pigs.

Do all pigs in a group of pigs have to be wild-type PRRSv-negative for vaccination to be effective, or can vaccination of the group still be effective if some pigs are wild-type PRRS virus positive?

No. All pigs do not have to be wild-type PRRSv-negative for the vaccination to be effective. Populations with a low PRRSv prevalence can be effectively vaccinated. Data suggests that vaccination recommendation rule applies: groups of pigs from positive, unstable farms with < 20% PRRS positive PCR pools are typically benefited from vaccination.

Does “therapeutic” vaccination reduce transmission risk when all, or a portion of a population of pigs are already wild-type PRRS virus exposed?

Yes. “Therapeutic” mass vaccination of PRRS positive or mixed-status populations of pigs with an MLV vaccine can reduce both the frequency and the duration of the transmission of wild-type PRRSv. Additionally, trials have demonstrated a significant reduction in the level and proportion of resident wild-type PRRSv over time.

Does “therapeutic” vaccination improve performance when all, or a portion of a population of pigs are already wild-type PRRS virus exposed?

Yes. Groups of pigs with < 20% PRRS-positive PCR pools are benefited from vaccination.

Will two doses of modified live vaccine 30 days apart for growing pig, replacement gilt or breeding animal vaccination provide better protection in relation to improved performance (due to reduced clinical problems) and reduction in transmission, compared to one dose, or does it make no difference?

Yes and No. Appropriately placed, one dose of Ingelvac PRRS MLV will provide significant protection to mitigate the consequences of infection and improve health and performance. Recent data from field-based studies suggest there are situations in the field, such as when pigs are sourced from PRRS-positive, unstable farms, where two doses of vaccine, four weeks apart, can have additional benefit over one dose. Current data would also suggest that the MLV vaccine, when used in a therapeutic population approach with a two-dose protocol, is more effective for reduction in transmission. In short, the two doses vaccination program is efficient when the pressure of infection in the region is high (i.e., high pig density with relatively high prevalence of PRRSv) and when wild-type PRRSv circulating in the region are of moderate to high virulence.

Vaccination

What are the vaccination recommendations for different PRRS statuses in farrow to finish systems?

Table 11. Vaccination recommendations for different PRRS statuses.

	PRRS status		Vaccinate breeding herd		PRRS status		Vaccinate growing pigs	
	Breeding herd		Sows (breeding, gestation, lactation)	Gilts	Shedding status (tested at weaning)*	Grow/Finish exposure		
1A	Positive unstable, High prevalence		Yes	Yes	⊕ High prevalence	⊕ Positive	Yes ¹	Control
1B	Positive unstable, Low prevalence		Yes	Yes	⊕ Low prevalence	⊕ Positive	Yes	
2	Positive stable		No	No	⊕ Uncertain	⊕ Positive	Yes/No	
2vx	Positive stable, W/Vaccination		Yes	Yes	⊕ Uncertain	⊕ Positive	Yes/No	
3	Provisional negative		No	No	⊖ Negative	⊕ Positive	Yes	
4	Negative		No	No	⊖ Negative	⊖ Negative	No	
								Elimination

*PRRS PCR assessment of pigs at weaning.

¹Studies demonstrate the benefit of vaccinating growing pigs when prrs prevalence is low (≤ 20% positive PCR pools). This decision should be made under the direct consultation of a veterinarian.

Economics





Is it correct that the economic impact of PRRS is higher in growing pigs?

That is correct. The misconception that PRRS is mainly impacting sows, is related to the less visual clinical impact in growing piglets. Whereas on the sow site abortion storms are very obvious, piglets infected with PRRS might show higher mortality rates and less weight gain. Checking production data will help to reveal the impact on sows and growing pigs.

Taken the whole-herd (sows and growing pigs), PRRSv is one of the most economically significant pathogens affecting the global swine industry. Economic losses from PRRS have been estimated at \$664 million annually in the United States, or \$1.8 million per day. That represents a profit loss of \$4.67 per growing pig, including losses due to poor performance and medication costs. (Holtkamp *et al.*, 2013).

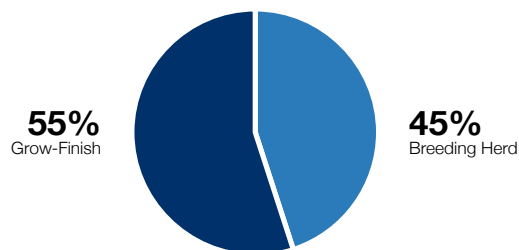
Figure 36. **Economic impact of PRRS.**

The total cost of productivity losses due to PRRSv in the US breeding and growing-pig herds was estimated at **US \$664 million** annually.

-  The total annual cost in breeding herds was **\$302.39 million** (45% of total cost).
-  The estimated annual cost in the growing-pig herd was **\$361.85 million** (55% of total cost).
-  The per-female cost was **\$114.71** per year.
-  On per-pig basis, PRRSv costs the US industry **\$4.67** for every pig marketed.

The majority of disease related costs are in growing piglets (55%).

Figure 37. **Total economic losses.**



Growing pigs infected with PRRSv have significantly worse average daily gain (ADG) and higher mortalities compared to pigs remaining PRRSv negative through market (Figure 26). The majority of economic losses are due to decreased revenue from marketing fewer pounds of pork.

However, many field studies report that the clinical disease is not always reported by farm staff. Thus, PRRSv causes a pseudo-subclinical disease in growing pigs, slowly but surely 'bleeding' producer's pockets.

Table 12. **Growing pig performance and profits of pigs infected with PRRS post-weaning compared to pigs remaining negative through marketing.**

Parameter	Negative weaning Through market	PRRSv infected post-weaning
Mortality rate	6.0%	7.4%
Average daily gain	1.56 lbs	1.53 lbs

What are my financial leverage points for most cost effective PRRS control?

To answer this question, Thomann *et al.*, 2020 evaluated the performance impact of PRRS on an endemic infected sow farm. As a second step the financial impact on different levels of control effectiveness and vaccine price were compared.

Result: The magnitude of benefits derived from vaccination was more susceptible to changes in vaccination effectiveness than to vaccine price changes.

Parameter	Negative farm	Infected farm	Disease effect
Return-to-oestrus rate	10.0%	13.5%	+3.5%
Abortion rate	2.0%	3.9%	+1.9%
Average piglets born alive per sow and litter	12.7%	11.4	-1.3
Pre-weaning mortality	11.0%	13.5%	+2.5%
Weight at weaning	6.0 Kg	5.5%	-0.5%
Days in nursery	45 days	50 days	+5 days
PRRS morbidity in weaners	–	20.0%	+20.0%
Mortality in weaners	3.0%	10.0%	+7.0%
Days in fattening	119 days	127 days	+8 days
PRRS morbidity in fatteners	–	20.0%	+20.0%
Mortality in fatteners	1.5%	3.0%	+1.5%



Thomann *et al.*, 2020

Economics

Thomann *et al.*, 2020

Sows: Double mass vaccination 4 weeks apart followed by a periodically mass vaccinated every 3 months. In-coming gilts are vaccinated twice during acclimatization.

Vaccination Effectiveness

Different levels of vaccination effectiveness were modeled. In this context, an assumed vaccination effectiveness of 80% would mean the following: If the baseline abortion rate in a PRRS negative farm is 2% and in a PRRSV-infected farm 3.9%, the absolute disease effect is +1.9%. Vaccination would reduce disease effects by 80% (-1.52%) and the abortion rate would persist at 2.38% after vaccination.

Vaccine price

The vaccine price was defined as the **price per dose (including labour) for the single vaccination of one sow**.

Table 13. Differences in annual gross margin (in €) between a PRRSV-infected farm without intervention and a PRRS-infected farm with mass vaccination of sows.

Vaccine price	Vaccination effectiveness				
	50%	60%	70%	80%	90%
0.75€	150,727	179,865	209,149	239,455	269,759
1.00€	149,581	178,387	208,127	238,336	268,272
1.25€	148,313	177,482	207,020	237,136	267,480
1.50€	147,525	176,312	205,959	235,820	266,435

Saving vaccination costs by 0.25€ per pig increases **the annual gross margin by 1.061€**.

Changing to a 10% more efficacious vaccine increases **the annual gross margin by 30.116€**.

Economics



Thomann *et al.*, 2020

Sows: Double mass vaccination 4 weeks apart followed by a periodically mass vaccinated every 3 months. In-coming gilts are vaccinated twice during acclimatization.

Piglets: vaccination between 2-3 week of live.

Vaccination Effectiveness

Vaccine price

The vaccine price was defined as the **price per dose (including labour) for the single vaccination of one sow**. For the whole herd strategy (sow and piglets), vaccination of a piglet would cost 80% of the price of sow vaccination.

Different levels of vaccination effectiveness were modeled. In this context, an assumed vaccination effectiveness of 80% would mean the following: If the baseline abortion rate in a PRRS negative farm is 2% and in a PRRSv-infected farm 3.9%, the absolute disease effect is +1.9%. Vaccination would reduce disease effects by 80% (-1.52%) and the abortion rate would persist at 2.38% after vaccination.

Table 14. Differences in annual gross margin (in €) between a PRRSv-infected farm without intervention and a PRRS-infected farm with mass vaccination of sows and piglets.

Vaccine price	Vaccination effectiveness				
	50%	60%	70%	80%	90%
0.75€	188,938	225,481	262,646	300,798	339,643
1.00€	182,137	218,858	255,985	293,787	332,766
1.25€	175,883	211,992	249,089	287,053	267,480
1.50€	169,563	205,473	242,660	280,265	319,222

Saving vaccination costs by 0.25€ per pig increases **the annual gross margin by 6.429€**.

Changing to a 10% more efficacious vaccines increases **the annual gross margin by 37.964€**.

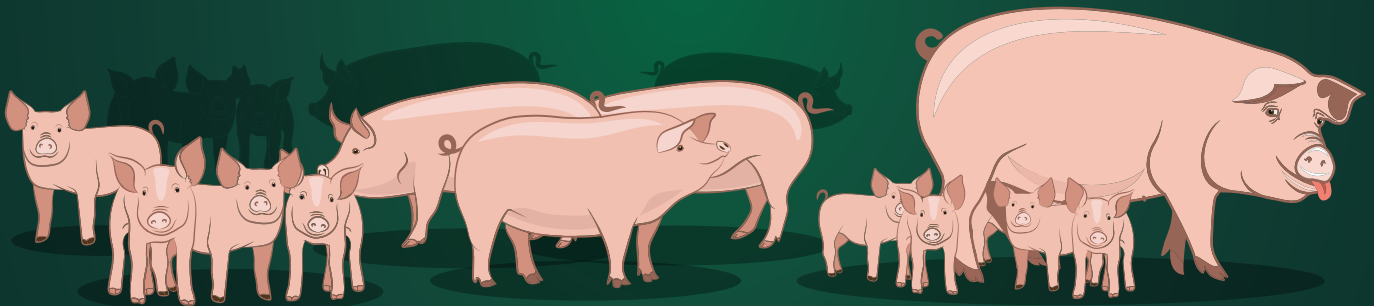
Economic impact of PRRS in sows and piglets

Where PRRS is costing you money

100%

Where you notice it

32%



COMBAT

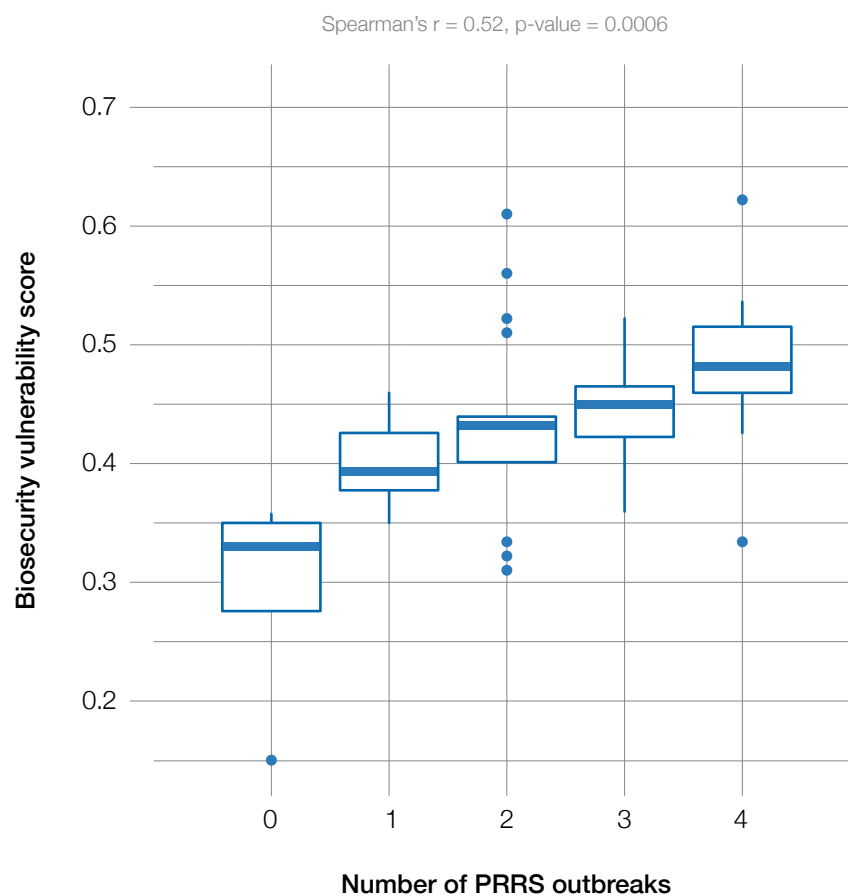
Why are Biosecurity improvements Crucial?

Biosecurity are all those measures applied to the farm to reduce the risk of introduction of pathogens in the farm (external biosecurity) and to minimize the spread of those pathogens within the farm (internal biosecurity). Having a good biosecurity has been associated with having fewer PRRS outbreaks, better performance indicators and lower antibiotic consumption.



G. Silva *et al.*, 2018

Figure 38. Biosecurity standards and PRRS outbreaks.



COMBAT is...

a free web based application will help to check and improve you PRRSV-related biosecurity by highlighting practices and procedures that can be reinforced. It provides:



1

Specific assessments

By selecting your production system, only relevant questions will be shown.

2

Immediate results

While conducting the assessment, get immediate results after each section (General, External, Transportation, Internal, Management)

3

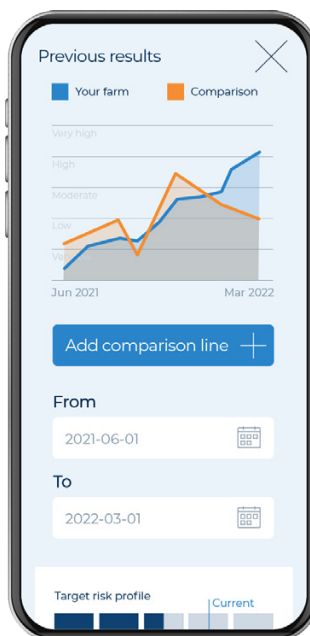
Actionable recommendations

A list of individual suggestions will help you to prioritize the actions to improve your biosecurity. Observe how your risk profile changes according to your selected actions.

4

Individual benchmarking

Evaluate your farm over time and compare to your own production system or to average country data.



Visit COMBAT

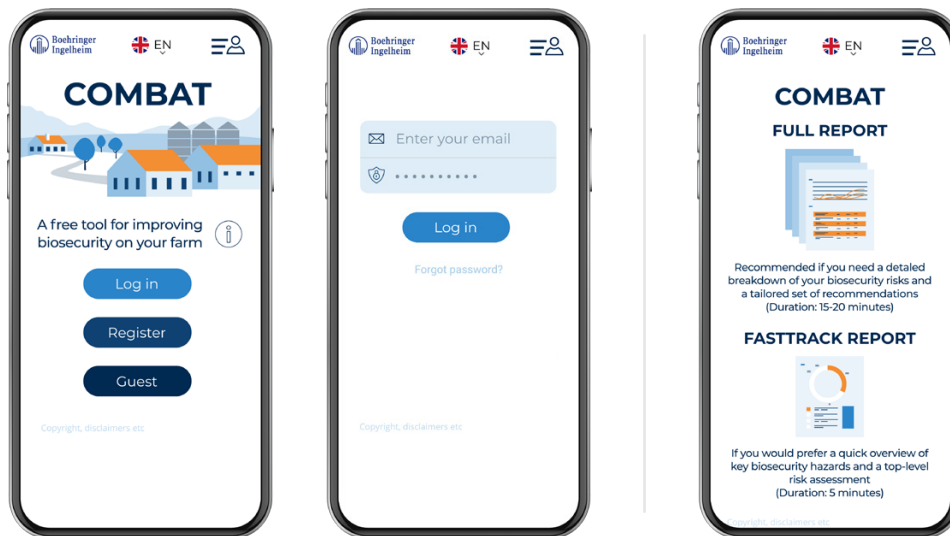
How to use combat: A pictured guide book

This chapter will explain how to apply COMBAT in a hands on scenario.

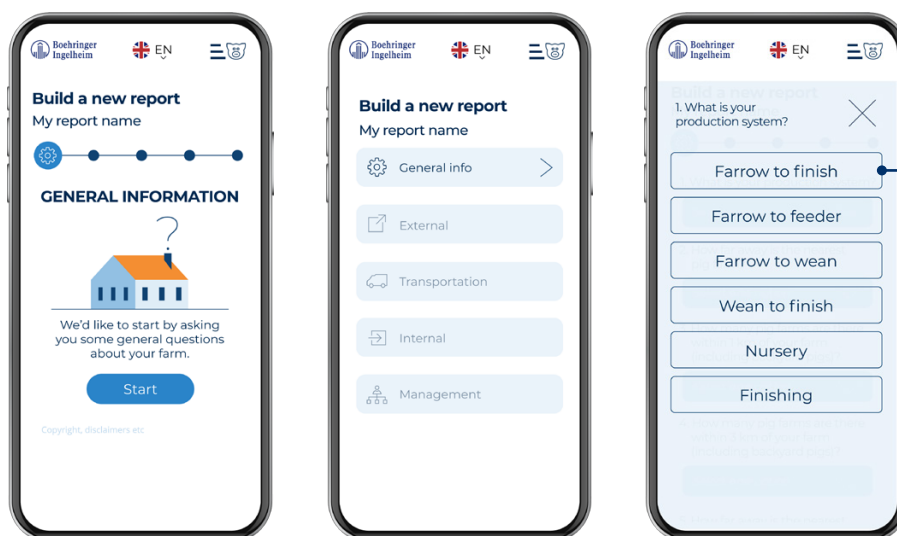
Farm settings:

- 500 Sows.
- Farrow to finish.
- Pig dense area.

Login and options selector



Build individual report



By selecting your production system, only relevant questions will be shown.

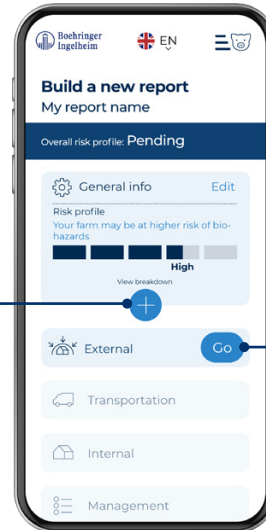
General info section

The general risk profile is based on your farm location and production type. In this case our farm was assessed as “High” due to the following factors:

- Farrow to finish.
- Small proximity to other pig farms with unknown PRRS status.
- Small proximity to major road with intense animal transportation.



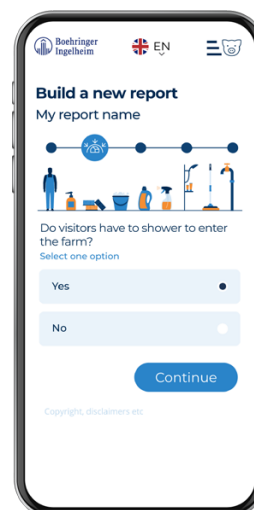
The breakdown option shows additional information on the rational behind the "high" biosecurity risk status in the "General info" Section.



After assessing the given Biosecurity status, we proceed with parameters that can easily be improved.

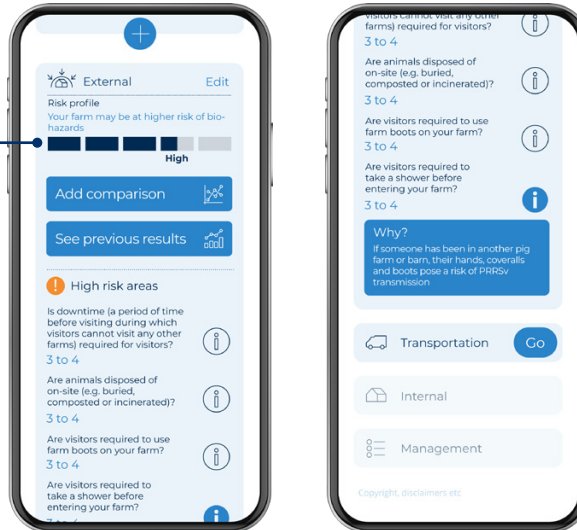
External section

Example Question: farm entry



Get stepwise feedback for each section

While we are proceeding with the Biosecurity assessment and are moving through the 5 sections (General, External, Transportation, Internal, Management) we receive stepwise feedback after each completed section.



Complete the remaining sections

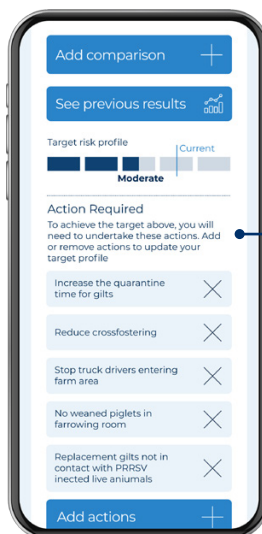
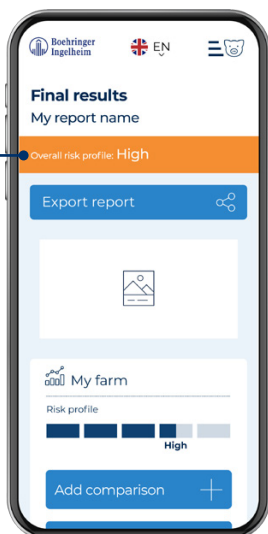


Verify that these are real data. In case you want to simply try out the tool, choose the latter option.

Results

You choose how to improve

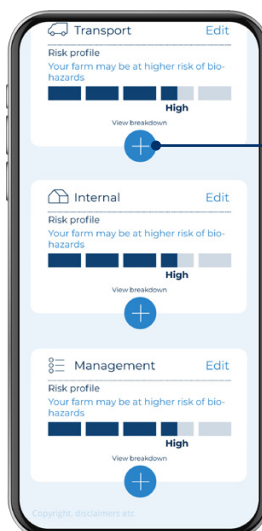
In our case scenario, the overall Biosecurity risk was evaluated as "High".



COMBAT is the only free biosecurity application that shows customized actions to reduce your farm risks, based on your previous assessment.

You can decide what actions are most feasible, as you know your farm best. Choose any action and see how your risk profile improves.

If you want to include specific actions, COMBAT will filter these for you and shows you the impact on your risk profile.

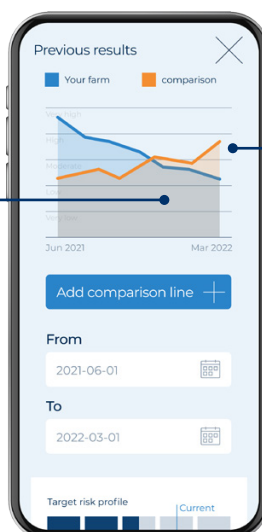
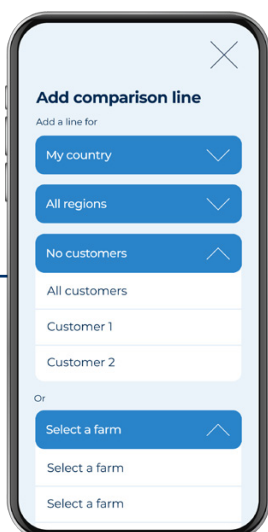


The breakdown menu shows you detailed information on each specific section.

And how to compare

A second unique feature of COMBAT is to benchmark your biosecurity status. As a comparison you can either select regional average of all similar production system (e.g. farrow to finish farms).

Besides that you can select a specific farm belonging to the same owner/production system to compare farm to farm (only in case you are the supervisor of this production system and have access to that farm data).



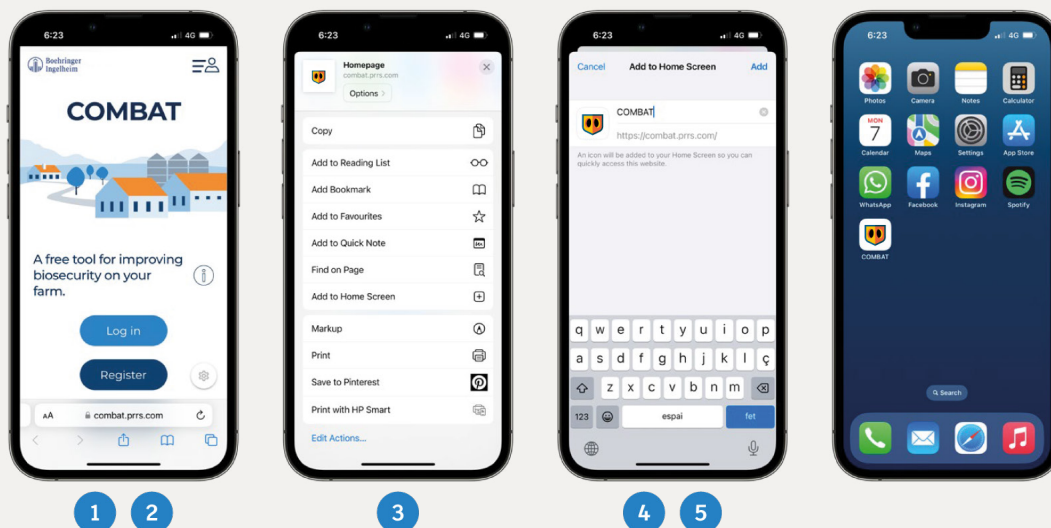
Keep track of your biosecurity status by repeating the survey on regular bases (e.g 3x/year).

Combat as a home screen bookmark

iOS

1. Launch **Safari**.
2. Navigate to "combat.prrs.com".
3. Tap the **Share** icon (the square with an arrow pointing out of it) at the bottom of the screen.
4. Scroll down to the list of actions and tap "**Add to Home Screen**".
5. Rename the link as **COMBAT**.
6. Tap "**Add**" in the top-right corner of the screen.

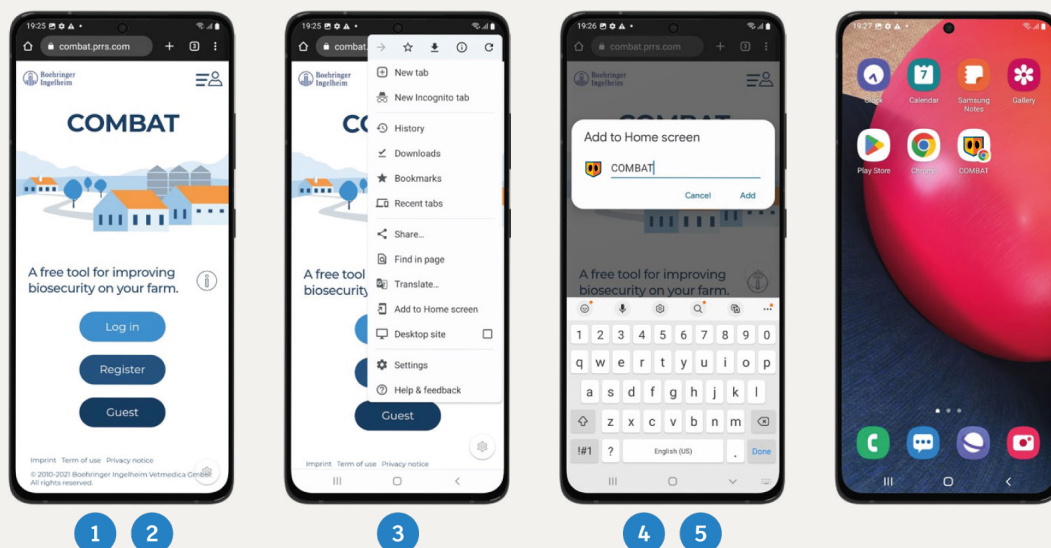
COMBAT, will appear as the last App Icon on your home screen.



Android

1. Launch **Chrome**.
2. Navigate to "combat.prrs.com".
3. Tap the **three-dot menu** on the top-right corner.
4. Tap "**Add to Home Screen**". It's toward the bottom of the menu, so you may have to scroll down to see it.
5. Rename the link as **COMBAT**.
6. Tap "**Add**".

COMBAT, will appear as the last App Icon on your home screen.



COMBAT – Updated biosecurity tool with 4 features

User friendly risk assessment, customized for your needs

Try it now for free



Now, with 4 features!



Visit COMBAT

4 are better than 1

- 1 Specific assessments
- 2 Immediate results
- 3 Actionable recommendations
- 4 Individual benchmarking

Crack the case

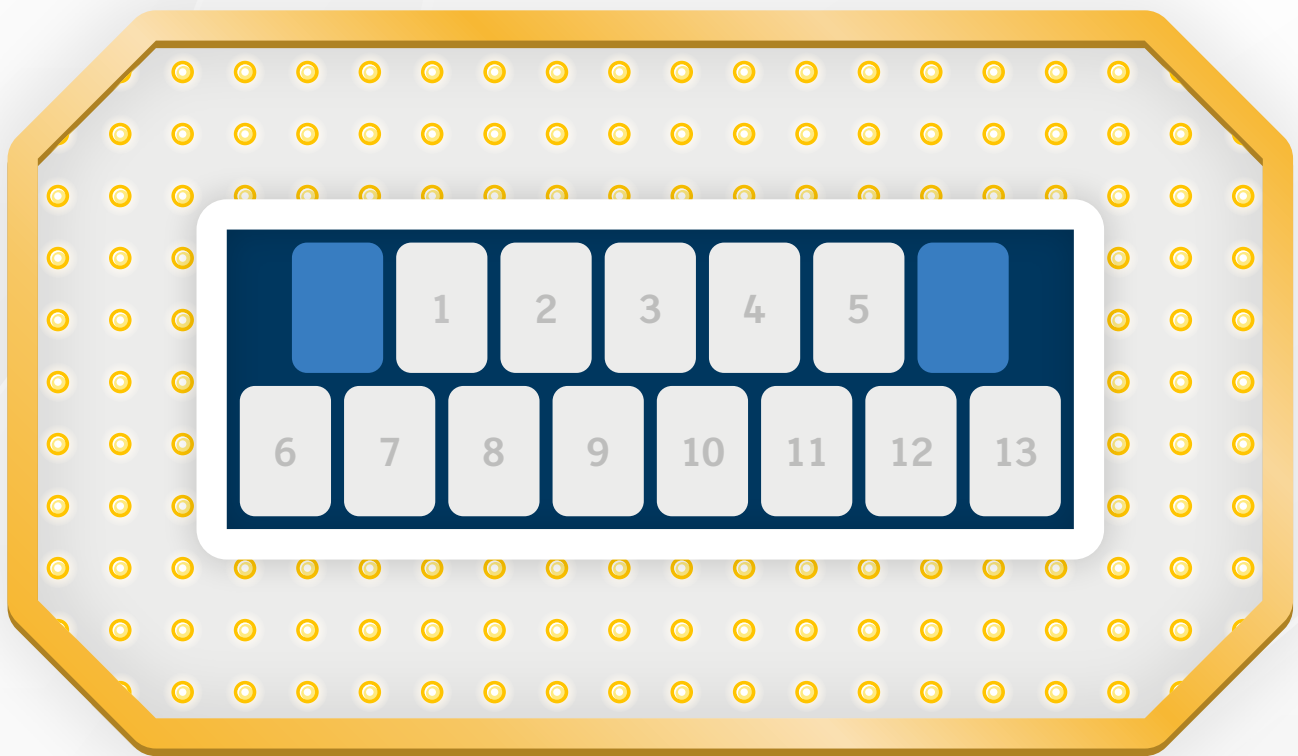
#1 Stabilize a breeding herd that broke with a Lineage 1 RFLP 1-4-4 PRRSv Type 2.

#2 Re-break or new introduction? Control PRRS in an infected sow farm with spike in abortions and increased number of sows off-feed.

#3 Entire Skill set needed! Stabilize a hyperprolific breeding herd with specific management challenges.

#4 Lost in Advice? A finishing farm located in a “biosecure” area broke with PRRS. Give targeted Biosecurity guidance and prioritize what to do.

Crack all four cases and reveal the hidden letters



Crack the Case #1

A previously PRRSv-stable (consistently weaning PRRSv-negative pigs) 6,000 Breeding herd broke with a Lineage 1 RFLP 1-4-4 PRRSv Type 2 (week 0).

Help the producer with a strategic PRRS control approach to decide on sample size and actions to be taken to reach stability again.



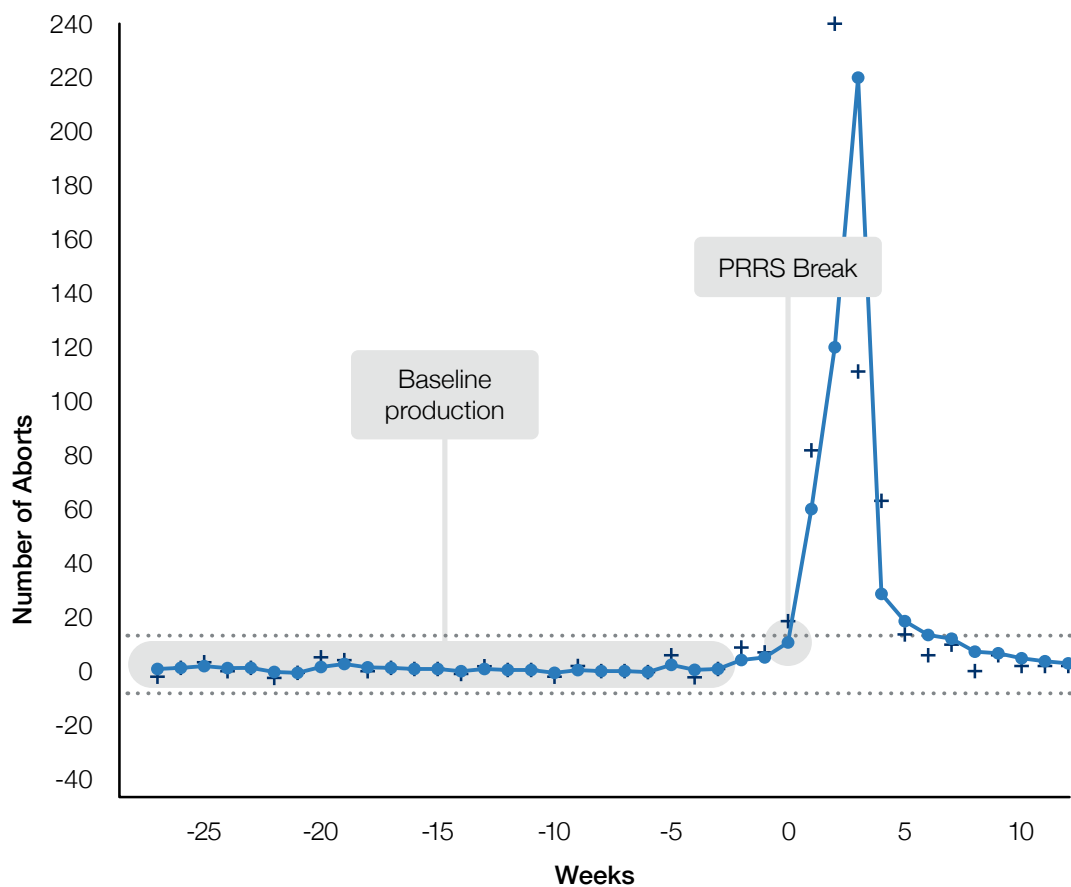
Crack the Case #1

Detection

There was an increase in abortions, from 3-4/week to 60 at week 0, 120 at week 1, and 220 at week 2. Also, there was a significant increase of sows off-feed in the gestation barn (n=90 week 0, 240 week 1).

As part of the 5 step process for systematic PRRS control, the goal of the farm (Step 1) was to control the virus and return to baseline production as soon as possible. But first assumptions of the responsible veterinarian that the farm broke with PRRS needed to be backed up with diagnostics.

Figure 39. Number of abortions in affected sow farm.



Question:

What can be done to confirm PRRSv infection in the herd to make sure that the spike in abortions and sows off-feed was attributed to the virus?

- A.** Collect 60 blood samples from gestating sows and test for PRRSv RNA by PCR.
- B.** Collect 60 blood samples from gestating sows and test for anti-PRRSv antibodies by ELISA.
- C.** Collect processing fluids from as many crates as possible within 1 week and test for PRRSv RNA by PCR.

Answer

It may take 2 weeks for most pigs seroconvert to PRRSv (i.e., test positive on ELISA), and therefore option “B” is the least preferable in this case. Option “A” works, but is labour, time, and cost intensive. Option “C” is the most practical, cheapest, and with higher herd sensitivity. In this case, processing fluids sampling took place at week 1, which included all processed litters from 5 farrowing rooms (56 crates each). The pooled processing fluids resulted PCR-positive with a Ct of 24, indicating PRRSv circulation in that population if castration is not performed, tongue tips fluids from stillbirth pigs are a great alternative to PF sampling. TT bags can contain 20-100 samples and can be frozen or refrigerated before or during transportation to the laboratory for testing by PCR. *To complete the final quiz, place a letter “C” on the box number 6.*

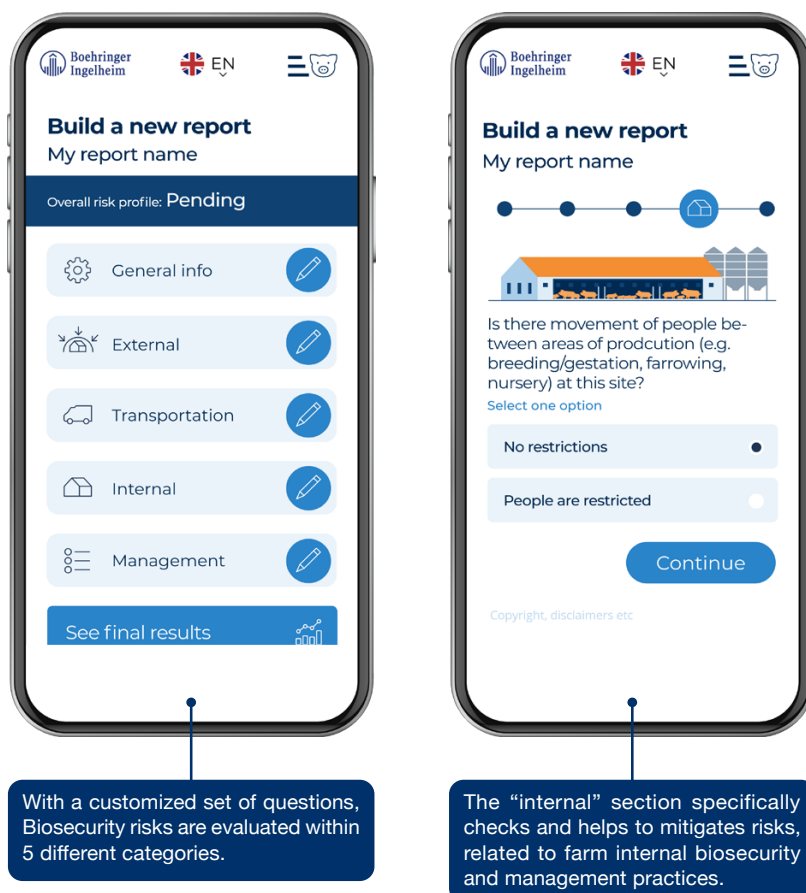
Crack the Case #1

Virus characterization

The processing fluid was submitted to whole genome sequencing of the PRRSV, which characterized the virus as wild-type PRRSV Type 2 with traces with other 2 wild-types previously reported, apparently a recombinant wild-type (Step 2: Determine current PRRS status).

Intervention

At weeks 3 and 5 post outbreak, the whole breeding herd (including sows and on site loaded gilt development unit) were subjected to mass exposure to Ingelvac PRRS MLV, and external gilt introduction was put on hold (i.e., herd closure). To reduce likelihood of viral transmission between crates and rooms, farm individual biomanagement practices were assessed and improved using the online tool COMBAT. (Step 3+4: Understand current constraints and develop solution options).



Question:

Was is a good choice to whole-herd expose both sows and gilts in this case?

- A.** Always good to expose only sows, not gilts.
- B.** In this case they could have relied on natural exposed and not used vaccine on sows nor gilts.
- C.** Mass exposure to both sows and gilts to MLV is the most efficient strategy to ascertain that all individuals will develop protective immunity, clearing viremia and shedding to both attenuated and wild-type viruses.

Answer

Option "C" is the correct answer. Not implementing herd closure, and not exposing the whole population of breeding females leads to extended time to negative and more severe production losses due to asynchronous exposure to the wild-type virus.

To complete the final quiz, place a letter "D" on the box number 12.

Crack the Case #1

Monitor implemented solution options (Step 5):

As part of Step 5 the success of implemented solutions was monitored. Weekly processing fluids continued to be collected and tested for PRRSv RNA by RT-PCR. At week 17 post outbreak (14 post intervention) processing fluids were still testing strong positive (consistently with Ct values below 30), and neonatal losses were still not back to baseline. This prompted further investigation of bio-management practices and pig flow within the herd.

It was verified that employees were still moving pigs between farrowing rooms, and hallways were not always decontaminated after pig movements. Strict bio-management practices were implemented, including stopping all piglet transfer between crates/rooms, keeping the floor and hallways clean and dry, and enforce personnel biosecurity during load-out of weaned pigs.



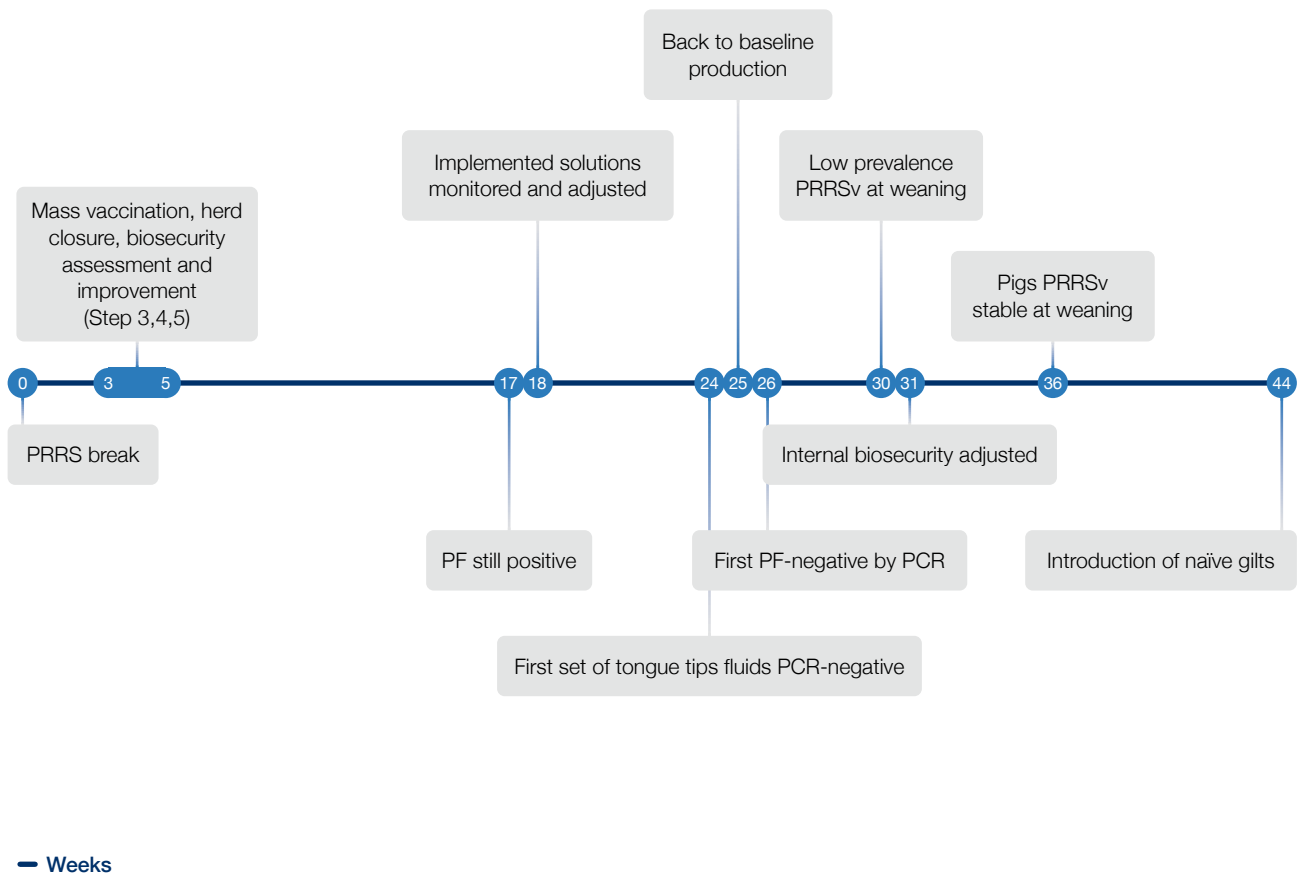
Crack the Case #1

At week 24 tongue tips (n=200) from stillbirth pigs tested negative by RT-PCR, indicating lack of PRRSv detection in the gestating sow population. Productivity wise (aborts, neonatal losses, preweaning mortality) the herd was back to normal. Between weeks 24-30 processing fluids also tested negative, confirming the consistent production of PCR-negative pigs.

This prompted the need to verify the status of weaning-age pigs. Thus, 20 family oral fluids samples were collected and tested in 4 pools of 5, from which 1 pool tested PCR-positive with a Ct of 31. Whole genome testing indicated 98% similarity with the original virus sequenced at week 1, indicating that external biosecurity was likely not an issue. Instead, this result suggests opportunities to avoid transmission of PRRSv from older to younger pigs, and between farrowing rooms.

Another meeting was held with farm employees explaining the importance of biocontainment practices, making it hard for the virus to move between time and space. After a few more adjustments, additional family oral fluids (FOF) tested PCR-negative starting week 36 onwards. Weekly processing fluid (PF) sampling also continued to test negative on PCR.

Figure 40. Timeline.



Question:

What if processing fluids testing was not an option due to not castrating pigs? What would be alternatives to cost-effective and reliable screening of the herd for PRRSv activity?

A. Tonsil scrapping of 30 suckling pigs per week.

B. Blood sampling 30 pigs per week.

C. 10 family oral fluids per week.

D. 10 Tongue tip samples

Answer:

Options “A” and “B” are both individual pig-based, and offer 95% confidence to detect virus at those sampled populations (week and room tested) when prevalence is 10% or higher. Family oral fluids is a population-based sampling approach (as processing fluids), where multiple pigs contribute to the sampling. 10 FOF samples is equivalent to about 90 serum samples, giving 95% confidence to detect at least one sample positive when prevalence is 3% or higher. Thus, option “C” is the correct one.

Answer “D” would also be applicable if samples size is at least 20 tongue tips samples per bag. Tongue tips fluids (TTF) are a great alternative, when 20+ samples from stillbirths and neonates are collected. The tongues can be collected daily and kept on the refrigerator or freezer for weekly submissions of 20-100 samples per bag.

To complete the final quiz, place a letter “K” on the box number 10.

Crack the Case #1

Recovery

The farm was declared stable at week 36 retrospectively at week 44 based on the recovery of clinical signs and PCR test results. Naïve gilts were introduced at week 44, and remained negative based on ELISA and PCR testing continuously until 2 months after introduction, when the herd was declared Provisional Negative.

Two years after that, following a complete herd roll over through about 50% annual replacement rate, the herd was declared Negative upon ELISA testing.

Congratulations, you cracked this case and gained a loyal customer. As a PRRS Expert you were invited to the "Meet the Expert" Podcast to share your practical knowledge.



Crack the Case #2

An endemic PRRSv infected sow farm had a spike in abortions and increased number of sows off-feed. Diagnostic testing confirms presence of wild-type PRRS virus.

The producer asks the veterinarian how to rule out a repeated outbreak (rebreak) with same virus that the farm had 1.5 years ago, versus new virus introduction.



Question 1:

Why is it important to determine if the virus being detected in the current outbreak is similar to the virus from the previous outbreak, versus unrelated (new) virus?

- A.** If it is the same virus, it means that internal biosecurity (biomanagement practices) should be the focus. If it is unrelated virus, it means that there are external biosecurity gaps that need remedy to prevent future outbreaks.
- B.** It doesn't matter the answer, the solutions are the same.
- C.** The producer is just curious. There is no way to answer this question.

Question 2:

What are tools that can be used to answer the question (old versus new PRRSv)?

- A.** ORF-5 sequencing, compare the most recent virus to the farm's library (i.e., past sequences) and to reference viruses from the local laboratory.
- B.** Whole-genome sequencing, compare the most recent virus to the farm's library (i.e., past sequences) and to reference viruses from the local laboratory.
- C.** ELISA testing, if the virus is new, the S/P ratios will be high.

Answer 1:

Option "A" is the correct answer. If it is the same virus, it means that internal biosecurity (biomanagement practices) should be the focus. If it is unrelated virus, it means that there are biosecurity gaps that need remedy to prevent future outbreaks.

To complete the final quiz, place a letter "C" on the box number 1.

Answer 2:

Option "A" or "B" are correct. With ORF5, it is expected that this portion of the genome changes about 1% per year. Thus, sequences $\geq 97\%$ similar provide strong evidence to support the same consensus sequence than the farm had before.

To complete the final quiz, place a letter "R" on the box number 7.

Crack the Case #2

Virus genomic testing and epidemiologic data provided evidence towards an outbreak with a completely different virus. The producer finds out that one employee was also working part time on a nearby finishing farm. That was the most likely source of infection. Several biosecurity improvements were made.

The farm is close to reaching stability, but still has about 10% of piglets still being weaned PRRSV-positive, and still don't perform as well in the grow-finish stage.

The producer wonders about piglet vaccination to improve piglet's health and is asking you for advice. Select your suggestion on the following page.



Question:

In this scenario, what are the benefits of vaccinating piglets?

- A.** MLV vaccine competes with wild-type infection. Exposing pigs to MLV develops partial heterologous protective immunity, helping to mitigate the clinical and production impact of the wild-type infection.
- B.** Less wild-type circulation leads to a decreased pressure of infection within the barn, and consequently to nearby farms.
- C.** MLV is a biocontainment tool, helping to decrease the magnitude and duration of shedding of wild-type virus.

Answer

All of the above are correct. The producer decides to implement vaccination of weaning-age pigs and observes a quick improvement on nursery and finishing performance of newly vaccinated flows. *To complete the final quiz, place a letter "E" on the box number 4.*

Crack the Case #3

A 600-head hyperprolific breeding herd with an on-site nursery in a separated building broke with PRRS.

Help the producer to stabilize the herd in the next 12 weeks and produce first negative piglets 30 weeks after the PRRS break.



Crack the Case #3

Directly after the PRRS break, the farm implemented a sow vaccination program 4 times per year to homogenize the herd immunity and to reduce the PRRS virus (PRRSv) dissemination in the farm. The farm also implemented a weaned piglet vaccination program.

To assess virus recirculation a PRRSv monitoring program was put in action by sampling processing fluids and serum at weaning. After the break, the veterinarian gave the farmer a protocol to stabilize the farm and produce negative piglets.

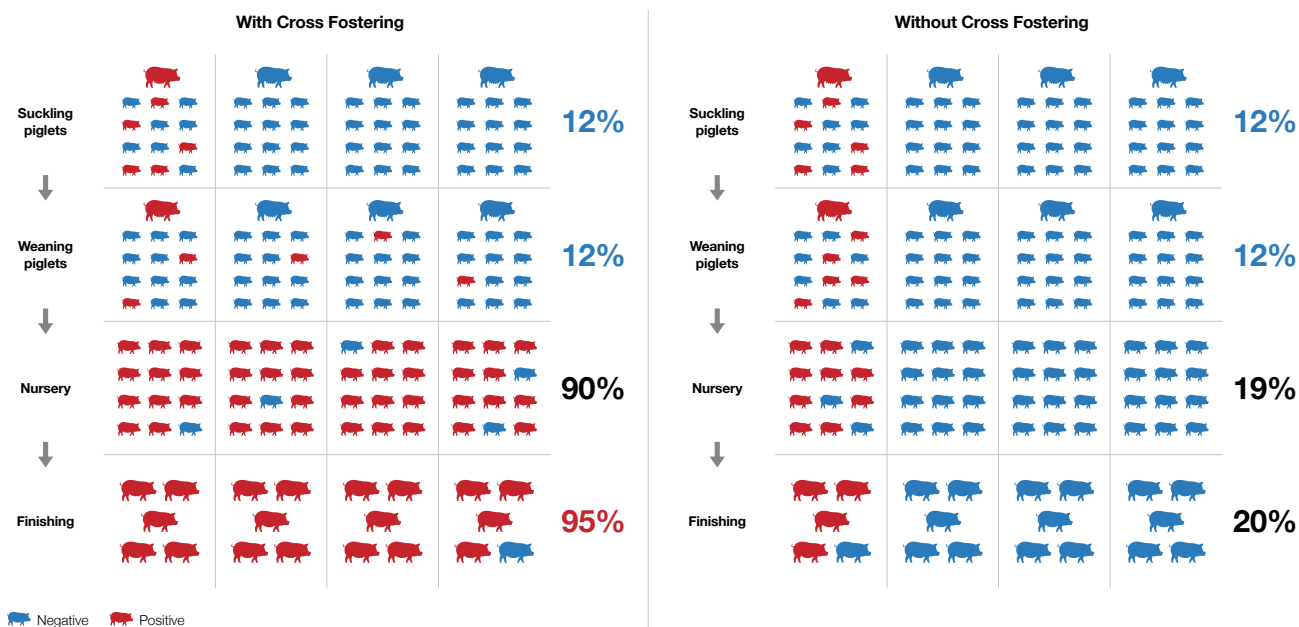
After a year the farm could not reach stability and processing fluids and serum from due-to-wean piglets was still positive by PCR. The virus infecting the piglets was the same virus that originated the outbreak based on ORF5 sequencing of positive samples pointing to internal biosecurity problems.

A new veterinarian from the company was assigned to the farm. As it was her first visit to the farm, she used COMBAT to get an initial biosecurity assessment with the focus on internal biosecurity.

As the sows give birth to high number of live piglets and the farmer wants to maximize their survivability, they do crossfostering. However, during the stabilization process they should reconsider their strategy.

Figure 41. Disease spreading with and without cross fostering. Courtesy: Prof. J. Dewulf.

See how diseases may spread in same prevalence scenarios with or without cross fostering.



Question 1:

How would you manage crossfostering?

- A. Pigs from different litters and age groups are comingled and moved to different rooms.
- B. Pigs from different litters and age groups are commingled and stay in same room.
- C. Pigs from different litters are mixed only with pigs of same age (less than one week difference).
- D. No crossfostering takes place.

Question 2:

Should we move nurse sows between batches?

- A. Yes.
- B. No.

Answer 1:

Answer "D". Ideally, we would not do crossfostering to avoid scattering the PRRSv and other pathogens in the farrowing room. However, working with highly prolific sows it is very difficult to not move piglets and the best recommendation would be to only crossfoster piglets of similar ages (less than one week) and in the same room (Answer C). As piglets have different states of immunity, we want to avoid that older piglets infect younger piglets. Pigs should most definitely NOT be cross-fostered to farrowing rooms with younger piglets. *To complete the final quiz, place a letter "C" on the box number 9.*

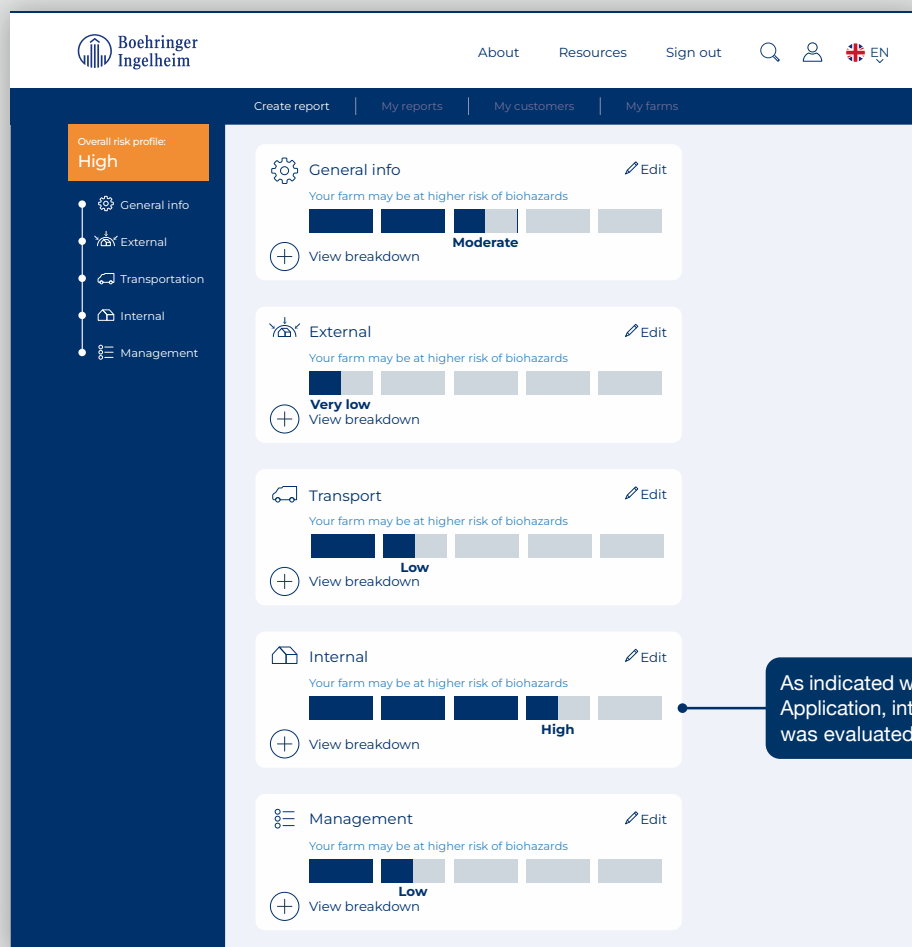
Answer 2:

Although beneficial for minimizing pre-weaning mortality and maximizing farm productivity, the use of nurse sows may facilitate the transmission of viruses (IAV and PRRSv) and bacteria to piglets prior to weaning. The use of nurse sows should be evaluated and stopped whilst the farm is unstable to avoid cross contamination.

To complete the final quiz, place a letter "A" on the box number 2.

Crack the Case #3

After the initial evaluation it was clear that some gaps existed in the internal biosecurity (biomanagement) of the farm. The movement of piglets and the use of nurse sows were evaluated and reduced to the minimum to avoid the spread of the virus.

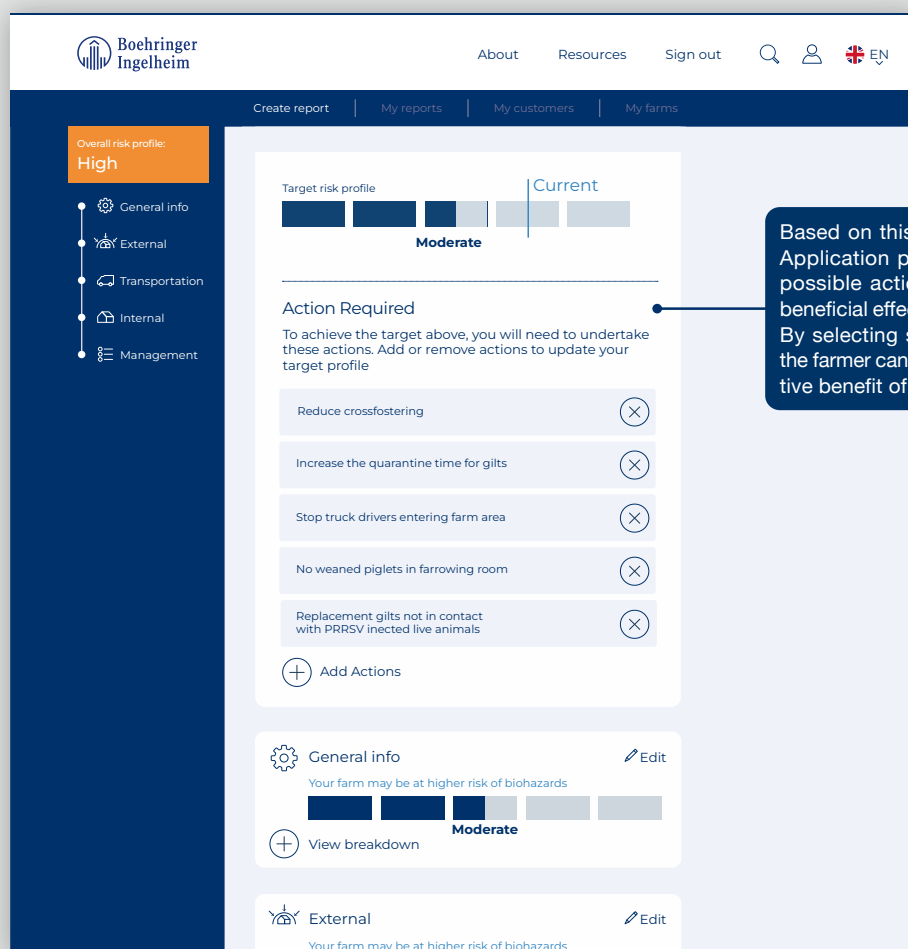


Crack the Case #3

Several weeks later the pigs were consistently born and weaned PRRSv negative according to processing fluids and serum sampling.

Some weeks after that the farmer called because PRRSv was still present in the nursery even though the piglets were weaned PRRSv negative.

In the visit to the farm, the veterinarian and the farmer reviewed and updated the biosecurity assessment done by COMBAT. As most of the issues related with the breeding herd were solved some other issues related to the way of managing the nursery and the movement of people were highlighted.



Question:

How would you run your nursery?

A. As continuous flow.

B. Pigs of different ages are mixed. When a room is emptied, underweight pigs are moved to a younger age group.

C. Pigs of different ages are mixed. When a room is emptied, all pigs leave.

D. All batches are kept intact from farrowing to slaughter without any mixing.

Answer

After piglets are weaned batch production should continue, and should be organized by site, barn or room. The risk of infection rapidly increases if a batch is not completely removed before new pigs are brought in. *To complete the final quiz, place a letter "E" on the box number 11.*

Crack the Case #3

The nursery barn was built before working with hyperprolific sows. The nursery had 8 rooms, 7 of the rooms are filled with piglets and one is empty, clean, and disinfected.

However, since started working with hyperprolific sows the flow of piglets has increased by 20% and nursery was never redimensionated. Because of the higher production, it was difficult to keep one specific room for each batch or even clean and disinfect the rooms between batches. The farmer considers adding extra spaces of nursery or selling one or two batches of piglets to reduce the risk of PRRSv recirculation.

During your visit, a worker that was working in the maternity was seen checking the different rooms of the nursery.



Question:

Would you advise to restrict people movement between areas of production (e.g. breeding / gestation, farrowing, nursery)?

A. Yes.

B. No.

Answer

Having restrictions in place regarding the movement of workers is the right choice. If workers need to move between different production areas, measures should be taken (e.g. boots and coveralls designated to each production area, clean hands or wear gloves and ensure the flow from high health, breeding unit, to low health, nursery or fattener). The reason is that after exposure to infected pigs, contaminated fomites (boots and coveralls) and hands can transmit PRRSv from infected to susceptible pigs. For this reason the movement of people should be reduced or biosecurity measures to reduce the spread of pathogens should be applied. *To complete the final quiz, place a letter "S" on the box number 3.*

Crack the Case #3

After the second visit the nursery was emptied temporarily to avoid the risk of having the virus introduced in the breeding herd again. During the empty period, the nursery was remodeled and expanded. A small changing room was added with dedicated coveralls and boots. The nursery will be visited every day before leaving.

Once resumed the use of the nursery the piglets were PRRSv negative until leaving to the fattening.

| Congratulations, you cracked this case and gained a loyal client.



Crack the Case #4

Two new 1,500-head finishing barns were built in a swine farm low density and hilly area. The farm is surrounded by forest. The farmer that owns the farm signed a contract with a company that decided to place PRRSv negative piglets there because it was located in a “biosecure” area because of the low pig density. However, the first three batches of pigs that were housed in the farm got infected by PRRSv.

The veterinarian assigned to this farm calls the farmer and asks him to do a quick biosecurity assessment using the fasttrack option from COMBAT. Help the veterinarian with the biosecurity check and help the farmer to prioritize what to do.



Question:

Should the building be surrounded by a perimeter fence?

- A.** None of the buildings should be surrounded by a perimeter fence.
- B.** Some of the buildings should be surrounded by a perimeter fence.
- C.** All of the buildings should be surrounded by a perimeter fence.

Answer

The farm's perimeter fence is the first line of defense that controls the movement of people, vehicles, and animals, especially wild pigs, from the traffic control area around a swine farm. Answer "C" is the one that will decrease the most of new PRRSv and other disease's introductions. In the example, the farm is in a forest area that can be home of different wildlife species that can act as vectors of PRRSv (wild boars) or other diseases. A complete perimeter fence should be built and kept it clean and clear of weeds and shrubs to avoid the entrance of external animals or unwanted visitors. *To complete the final quiz, place a "!" on the box number 13.*

Crack the Case #4

Because the farm was built in a low-density area, the farmer thought that could save some money by not building a fence surrounding the premises.

After the biosecurity assessment and the discussion with the veterinarian it was clear that a fence was a 'must' to avoid the entrance of wildlife risky animals and control external vehicles.



Question:

How frequently should the vehicles used to transport animals to market or collection point be cleaned?

A. Never.

B. At least once per 20 loads.

C. At least once per 10 loads.

D. Between every load.

Answer

Answer "D". PRRSv can be present in trucks that have not been washed properly or the disinfectant was not adequate. Trucks should be cleaned after every load. The presence of the virus in future loads can increase the risk of transmission to the newly loaded animals and to the animals in the farm. *To complete the final quiz, place a letter "S" on the box number 5.*

Crack the Case #4

It was clear that during the assessment some of the trucks did not come clean specially those trucks that were loading the final cuts. The veterinarian contacted the truck company to ask for clean and disinfected trucks given that the farm is being used to raise negative pigs and the pig company is very interested in keeping the farm that way in case they need to use it one day to raise gilts.

The farmer corroborates that truck drivers, in order to help, sometimes enter the hall-way to push the pigs to the truck and fasten the load. The veterinarian will propose to build a stage loading or a bottomed-open fence that only allows the movement of pigs.



Question:

Where should dead animals be collected from?

- A. Inside the farm perimeter.
- B. Less than 500m (0.3 miles) from the farm/site.
- C. More than 500m (0.3 miles) from the herd, but internal and third-party vehicles cross the rendering vehicle route.
- D. More than 1km (0.6 miles) from the herd, but internal and third-party vehicles cross the rendering vehicle route.
- E. More than 1km (0.6 miles) from the herd, but internal and third-party vehicles never cross the rendering vehicle route.
- F. On-site disposal (incinerator, hydrolisis or burial).

Answer

Answer “F” (if allowed in your country, otherwise Answer “E”). On site disposal methods are the ones with the lowest risk of PRRSv introduction. However, not all of them are available in each country. The most spread used method of cadaver disposal is through rendering. There is a scarcity of information of the risk of rendering trucks for PRRSv spread. However, in the available literature it was found that farms with the rendering truck entering the premises had been 7 times more associated to being positive compared to those who did not allow the truck entering the premises. Ideally, the dead animals or disposal containers for pick up should be placed external to the fenced perimeter as far as possible from the farm buildings. *To complete the final quiz, place a letter “A” on the box number 8.*

Crack the Case #4

Currently, the farm is relying on a rendering company to get rid of the cadavers. Incinerating is not allowed in the country and the farmer never heard of hydrolysis before. In order to facilitate the movement of dead pigs the container for cadaver disposal was placed between the two buildings at the back of the farm. The rendering truck had to drive around the farm close to the buildings to access the container and turn around.

Following the veterinarian instructions, the container will be placed outside the fence (once is built) with different access sides for the farmer and the truck so the risk of cross contamination will be minimized.

Congratulations!

Based on your recommendations, biosecurity policies were improved and subsequent batches of pigs remained negative.



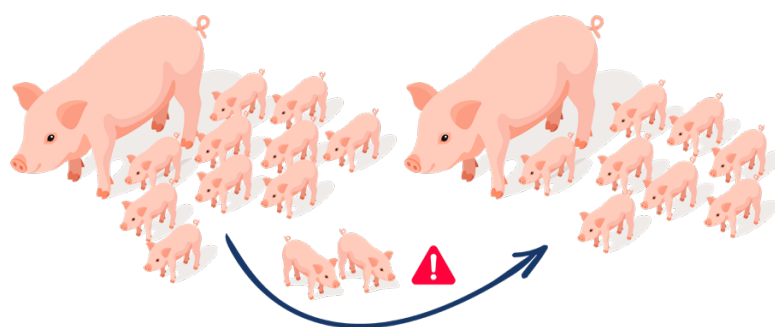
Swiss Cheese Model

Basic Biosecurity Guidelines to reduce movement/transmission of virus within and between groups or populations of pigs.

Rule #1: When cross-fostering, move pigs only when necessary

At the time of farrowing, it is not unusual for the sows within a farrowing group to have a high number of piglets per litter, and after considering factors such as the number of functional teats and teat conformation, farrowing technicians commonly carry out a process called “cross-fostering”. This movement of piglets to a foster sow of the same group is also referred as “litter equalisation” it may actually negatively impact health stability **so it should always be minimized, especially for PRRS positive farms!**

- Since all **piglets are not born with the same immune status (PRRS)**, and have not received the same colostrum, **any piglet movement between litters has the potential to spread pathogens (PRRSv) within the lactation room.**¹
- Any piglet movement between litters has the potential to disrupt the milk production of the sow causing weight loss and affecting the well-being of the moved piglets and the entire litter, **so avoid unnecessary movements.**²
- It has been shown that **piglets that were cross-fostered once were 11.69 times more likely to die** and were at higher risk of pericarditis and heart condemnation compared with pigs that were not cross-fostered ($p < 0.05$).³








Rule #1

When cross-fostering,
move pigs only when necessary



¹ Garrido-Mantilla, J., Culhane, M., Torremorell, M., 2019. Experimental transmission of influenza A virus and porcine reproductive and respiratory syndrome virus from nurse sows to adopted pigs during lactation. 50th Annual Meeting of the American Association of Swine Veterinarians Orlando, pp. 54 March 9-12, 2019.

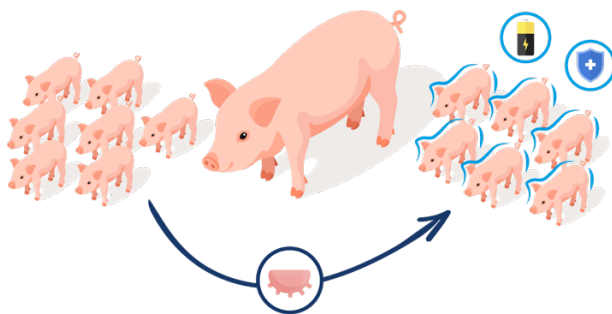
² Alexopoulos JG, Lines DS, Hallett S, Plush KJ., 2018. A Review of Success Factors for Piglet Fostering in Lactation. *Animals (Basel)*. 2018 Mar 9;8(3):38. doi: 10.3390/ani8030038. PMID: 29522470; PMCID: PMC5867526.

³ Calderón Díaz, J.A., García Manzanilla, E., Diana, A., Boyle, L.A., 2018. Cross-fostering implications for pig mortality, welfare and performance *Front. Vet. Sci.*, 5 (2018), 10.3389/fvets.2018.00123.

Rule #2: No cross-fostering later than 48 hours

After farrowing, providing adequate colostrum intake while minimizing cross-fostering at the same time is an industry challenge. Colostrum intake is one of the main determinants of piglet survival since **it provides the essential energy and immunity that every piglet needs in early life**. Colostrum from the birth sow also helps to maximize the quality of immunity. But the pressure to cross-foster early can be high due to reasons such as increased sow prolificacy, inadequate functional teat numbers, and disease instability. Since early cross-fostering can result in the variability of the quality and volume of colostrum intake of piglets within the litter **it is important to delay the process as long as possible**. At the same time, **the cross fostering that is absolutely necessary must not occur too late in lactation** in order to avoid litter disruption and the associated negative impacts.

- **Piglets are born immunologically naïve** as the sow is unable to transfer antibodies in utero to the piglets via placenta, **so antibody transfer from colostrum is crucial for adequate immune function** in early life. Immunoglobulin (Ig) G is the most predominant in colostrum and its concentration decreases dramatically during the first 24-30 hours of life.¹ Some cellular immunity cells are incompatible to pigs of other litters. In other words, full transfer of maternally-derived immunity only happens to pigs born to their dam. Transferring piglets to suck on other sows can impair the intake of protective immunity against various pathogens.
- Piglet ability to absorb antibodies from colostrum **decrease rapidly after 6 hours from the first feeding** due to a permeability decrease of large proteins through gut membranes.



¹ Alexopoulos JG, Lines DS, Hallett S, Plush KJ. A Review of Success Factors for Piglet Fostering in Lactation. *Animals (Basel)*. 2018 Mar 9;8(3):38. doi: 10.3390/ani8030038. PMID: 29522470; PMCID: PMC5867526.2 Tuboly S., Bernath S., Glavits R.K., Medveczky I. Intestinal absorption of colostral lymphoid cells in newborn pigs. *Vet. Immunol. Immunopathol.* 1988;20:75-85. doi: 10.1016/0165-2427(88)90027-X.

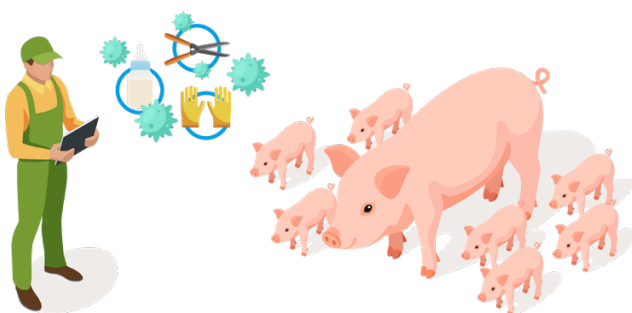
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Rule #2
No cross-fostering later than 48 hours


Rule #3: Keep piglets in the farrowing pen and avoid handling to minimise the spread of disease

After the farrowing event, there are several management practices that require the handling of piglets. Examples include piglet processing (e.g. clipping teeth, tail docking, umbilical cord management, iron administration, and castration), split suckling using warm boxes, and cross-fostering. To perform all of these processes, it is not uncommon for the farrowing technicians to step into the farrowing pens and share common tools to hold and treat piglets from each litter in order to ensure the most efficient work processes. However, **all of these management practices can facilitate the spread of pathogens such as PRRSv between litters and rooms.** Keeping the piglets in their own pen and minimising the number of handling processes that share tools and spaces is critical for disease management.

- Possible indirect routes of PRRS virus transmission include urine, blood, saliva and faeces from infected animals to susceptible ones. Warming boxes and processing carts when **shared within the same room can serve as fomites to transmit the virus between litters.**¹
- **Avoid stepping into the farrowing crates of each sow.** It has been demonstrated that boots and coveralls can serve as fomites for the PRRS virus from infected to susceptible animals.¹






Rule #3

Keep piglets in the farrowing pen and avoid handling to minimise the spread of disease

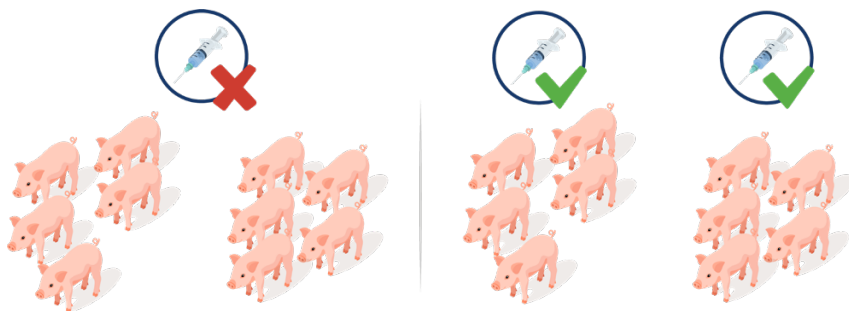


¹ Otake S, Dee SA, Rossow KD, Deen J, Joo HS, Molitor TW, and Pijoan C. Transmission of porcine reproductive and respiratory syndrome virus by fomites (boots and coveralls). *Swine Health Prod* 2002. 10(2): 5965.

Rule #4: Change needles between litters

Syringes and needles are used to administer injectable antibiotics and analgesics (i.e. pain killers) as well as other therapeutic products such as iron and vitamins. In the swine industry, it is common to use the same needle to inject the same product into different animals. **The practice of sharing needles between piglets during the lactation period can play a significant role in the transmission of infectious diseases, such as PRRS.**

- At the peak of viremia, infected animals have a viral load of at least 10^3 - 10^4 TCID₅₀/L.
- Assuming a minimum infectious dose of 10^1 - 10^2 TCID₅₀ through a percutaneous (i.e. injectable) exposure, a simple drop of blood (i.e. 1-10 µL) could transmit a sufficient amount of virus between animals.¹ Even if a needle change is performed between litters, an additional measure to minimize within litter spread would be to inject small and rough-haired animals last.
- **Bloodborne transmission of the PRRS virus has been demonstrated** in controlled field studies using both the same needle and needle-free injection devices.^{2,3}



¹ Duan X, Nauwynck HJ, Pensaert MB (1997) Virus quantification and identification of cellular targets in the lungs and lymphoid tissues of pigs at different time intervals after inoculation with porcine reproductive and respiratory syndrome virus (PRRSv). *Vet Microbiol* 56:9–19.

² Otake S, Dee SA, Rossow KD, Joo HS, Deen J, Molitor TW, Pijoan C. Transmission of porcine reproductive and respiratory syndrome virus by needles. *Vet Rec.* 2002 Jan 26;150(4):114-5. PMID: 11838995.

³ Baker SR, Mondaca E, Polson D, et al., Evaluation of a needle free injection device to prevent hematogenous transmission of porcine reproductive and respiratory syndrome virus. *J Swine Health Prod.* 2012 ; 20(3):123-128

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Rule #4
Change needles between litters

Rule #5: Do not move sick piglets

During the lactation period, management practices and factors impacting stress levels and disease status **have the potential to influence milk production** and, with that, piglets and litter performance. Therefore, litters and piglets within litters, do not always show the same growth performance. This reality is common for PRRS positive farms. To fix this issue, producers tend to practice cross-fostering of piglets that are falling behind, when compared to their pen mates without considering that **the disease transmission risk could be higher than the potential improvement in growth performance**.

- The movement of sick piglets and runts, often referred to as ‘fall-back piglets’, **increases the probability of pathogen transmission between litters** due to animal-to-animal contact with different immune status for PRRS and other pathogens.¹
- Management practices at the farrowing, such the use of nurse sows, has been demonstrated **to facilitate PRRS virus transmission** to piglets from the sow and to sow from sick piglets.²








Rule #5

Do not move sick piglets



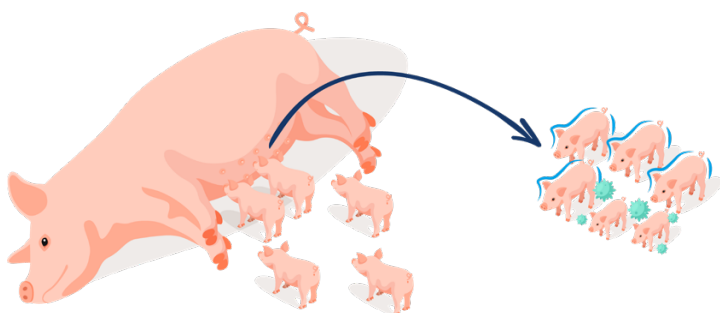
¹ D. Maes, J. Segales, T. Meyns, M. Sibila, M. Pieters, et al., Control of infections in pigs. *Vet. Microbiology*, Elsevier, 2009, 126 (4), pp.297. [ff10.1016/j.vetmic.2007.09.008](https://doi.org/10.1016/j.vetmic.2007.09.008). [ff10.1016/j.vetmic.2007.09.008](https://doi.org/10.1016/j.vetmic.2007.09.008).

² Garrido-Mantilla, J., Culhane, M.R. & Torremorell, M. Transmission of influenza A virus and porcine reproductive and respiratory syndrome virus using a novel nurse sow model: a proof of concept. *Vet Res* 51, 42 (2020). <https://doi.org/10.1186/s13567-020-00765-1>.

Rule #6: Wean all piglets from the same farrowing group at the same time, and do not allow any weaned piglets to remain in farrowing rooms

Correct growing pig flow starts in the farrowing room when piglets, that were born into the same farrowing group, are weaned away from the farrowing rooms at the same time to create a batch of growing animals with a similar age. **Pig flow errors at the time of weaning can compromise overall health stability.** Examples of these errors include cross-fostering underweight wean-aged piglets onto younger litters (i.e. 'hold back piglets') for quality/weight improvement and allowing piglets that have been weaned from their sow to stay in the farrowing crate without a sow. Despite the potential benefits, **these practices put the health status of the farm at risk.**

- When the milk supply is terminated at weaning, maternal antibody levels decline which **leaves the piglet vulnerable to infections** that can drive changes in the dynamics of disease transmission within the population.^{1,2} Therefore, **if weaned piglets remain in the farrowing rooms, they become a potential source of pathogens for disease transmission** to other litters and sows that are on their way back to the breeding area.
- The movement of older hold-back piglets from one batch to the next **increases the probability of disease transmission between animals of different ages** via direct and indirect contacts between piglets with different immune statuses for PRRS and other pathogens.³



¹ Kraft C, Hennies R, Dreckmann K, Noguera M, Rathkjen PH, Gassel M, et al., (2019) Evaluation of PRRSv specific, maternally derived and induced immune response in Ingelvac PRRSFLEX EU vaccinated piglets in the presence of maternally transferred immunity. PLoS ONE 14 (10): e0223060. <https://doi.org/10.1371/journal.pone.0223060>.

² Geldhof MF, Van Breedam W, De Jong E, Lopez Rodriguez A, Kamiychuk UU, Vanhee M, et al., Antibody response and maternal immunity upon boosting PRRSv-immune sows with experimental farm-specific and commercial PRRSv vaccines. Vet Microbiol. 2013;167(3-4):260-71. Epub 2013/09/18. pmid:24041768.

³ D. Maes, J. Segales, T. Meyns, M. Sibila, M. Pieters, et al., Control of infections in pigs. Vet. Microbiology, Elsevier, 2009, 126 (4), pp.297. #10.1016/j.vetmic.2007.09.008ff. fthal-00532322f

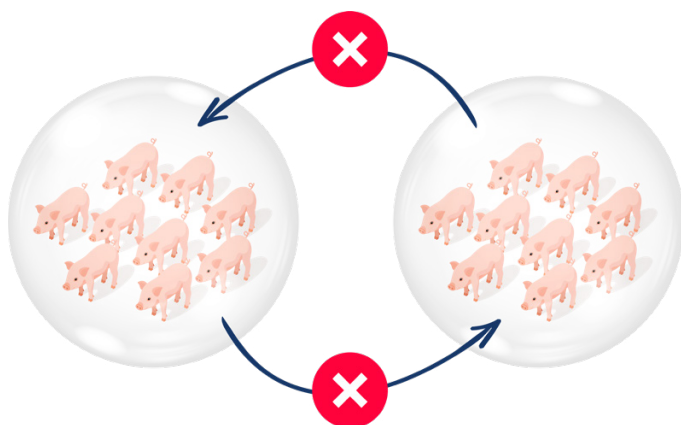
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Rule #6
Wean all piglets from the same farrowing group at the same time, and do not allow any weaned piglets to remain in farrowing rooms

Rule #7: Strict batch production (all in/all out)

All-in/all-out systems keep pigs together within batches as they move through the different phases of production. Each group is considered to be an intact unit and, once the group has moved forward, the facility or room is completely emptied and cleaned for the next group. Although the principle of all in/all out pig flow seems simple, **it is one of the most difficult rules to implement** due to variation in production parameters such as: pigs per batch, weight range, and growth rates. When the causes of variation are not addressed, managers are forced to compensate by breaking the all in/all out rule.

- Cleaning and disinfection strategies between batches is probably **the most important internal biosecurity measure** to break the infectious cycle of pathogens from one production batch to the next.¹
- Mixing or sorting pigs is a source of stress to the animals and **it increases the probability of disease transmission due to differences in the immune status** for PRRS virus and other pathogens.²
- **Do not share needles, equipment, personnel and protective equipment between batches** (unless cleaned and disinfected) since it could increase indirect transmission of the PRRS virus.³








Rule #7

Strict batch production (all in/all out)



¹ Clark, L., Freeman, M., Scheidt, A., Knox, K., 1991. Investigating the transmission of 538 *Mycoplasma hyopneumoniae* in a swine herd with enzootic pneumonia. *Vet. Med.* 86, 539-543-550.

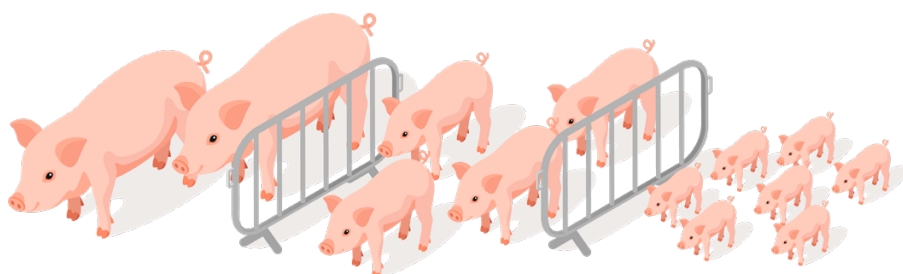
² D. Maes, J. Segales, T. Meyns, M. Sibila, M. Pieters, et al., Control of infections in pigs. *Vet. Microbiology*, Elsevier, 2009, 126 (4), pp.297. [ff10.1016/j.vetmic.2007.09.008ff](https://doi.org/10.1016/j.vetmic.2007.09.008). [ff10.1016/j.vetmic.2007.09.008ff](https://doi.org/10.1016/j.vetmic.2007.09.008).

³ Rathkjen and Dall Control and eradication of porcine reproductive and respiratory syndrome virus type 2 using a modified-live type 2 vaccine in combination with a load, close, homogenise model: an area elimination study. *Acta Vet Scand* (2017) 59:4 DOI 10.1186/s13028-016-0270-z.

Rule #8: No contact between different age groups

In well designed production systems, correctly dimensioned facilities allow producers to house each production batch in separate buildings/rooms (i.e. independent air spaces) at the correct pig density to maintain proper All In/All Out (AI/AO) pig flow management. **This strategy ensures no contact between pigs of different ages.** However, good AI/AO flow can be challenged due to factors such as significant variation in batch size or the presence of disease. These examples can result in differences in pig growth and quality which can force the need to mix animals from different batches (i.e. different ages) in the same air space.

- Mixing pigs of different age groups at any stage of the production is a **risky practice**, as it can bring pathogens to a susceptible population due to differences in the immune status for the PRRS virus.^{1,2}
- Nathues *et al.*, estimated that contact between fattening pigs of different ages during restocking of compartments increased by **13 times the risk for respiratory disease occurrence** in enzootic pneumonia positive herds.³



¹ D. Maes, J. Segales, T. Meyns, M. Sibila, M. Pieters, et al., Control of infections in pigs. *Vet. Microbiology*, Elsevier, 2009, 126 (4), pp.297. #10.1016/j.vetmic.2007.09.008ff. f#hal-00532322f.

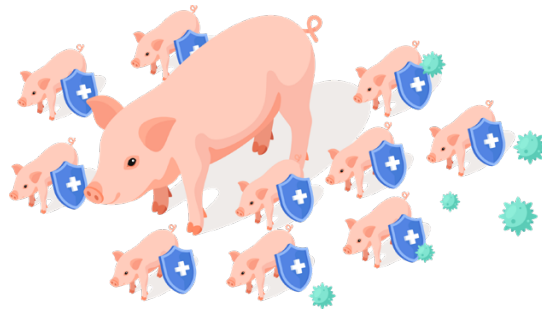
² Filippitzi ME, Brinch Kruse A, Postma M, et al., Review of transmission routes of 24 infectious diseases preventable by biosecurity measures and comparison of the implementation of these measures in pig herds in six European countries. *Transbound Emerg Dis.* 2017;00:1–18. <https://doi.org/10.1111/tbed.12758>.


³ Nathues H, Chang YM, Wieland B, Rechter G, Spergser J, Rosengarten R, Kreienbrock L, Grosse Beilage E. Herd-level risk factors for the seropositivity to *Mycoplasma hyopneumoniae* and the occurrence of enzootic pneumonia among fattening pigs in areas of endemic infection and high pig density. *Transbound Emerg Dis*. 2014 Aug;61(4):316-28. doi: 10.1111/tbed.12033. Epub 2012 Dec 2. PMID: 23199301.

Rule #9: No contact between pigs less than six months of age and sows

After farrowing, piglets receive powerful immunity from the sow via colostrum and milk that **makes them immune to most of the pathogens to which the sow has been exposed**. However, this immune status starts waning in the piglets right after weaning with the removal of the milk supply. After weaning, piglets must use their own active immunity to protect against the infectious challenges they meet. Several factors can inhibit or challenge the growing pig's immune response such as changes in nutrition, stress due to poor management practices, and commingling with other pig populations that are a source of infectious challenges. **Ensuring the separation of growing pig batches from the sow herd protects the sow herd from potential disease challenges.**

- Several studies have demonstrated that **younger animals have significantly longer viremia**, and higher viral loads in lymph nodes, and lungs.^{1,2}
- Sow farms with a growing pig population (e.g. wean to finish gilt development units attached to a sow unit) that is not properly isolated from the sows **are more likely to show a longer persistence of PRRSv** infection following an outbreak.³








Rule #9
No contact between pigs less than six months of age and sows

¹ Klinge KL, Vaughn EM, Roof MB, Bautista EM, Murtaugh MP. Age-dependent resistance to porcine reproductive and respiratory syndrome virus replication in swine. *Vet J.* 2009;6:177. doi: 10.1186/1743-422X-6-177.

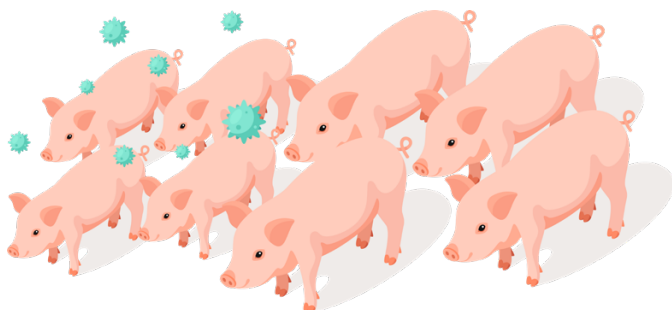
² Cho JG, Dee SA, Deen J, Trincado C, Fano E, Jiang Y, Faaborg K, Murtaugh MP, Guedes A, Collins JE, Joo HS. The impact of animal age, bacterial coinfection, and isolate pathogenicity on the shedding of porcine reproductive and respiratory syndrome virus in aerosols from experimentally infected pigs. *Can J Vet Res.* 2006;70:297-301

³ Evans CM, Medley GF, Creasey SJ, Green LE. A stochastic mathematical model of the within-herd transmission dynamics of porcine reproductive and respiratory syndrome virus (PRRSv): fade-out and persistence. *Prev Vet Med.* 2010;93:248-257. doi: 10.1016/j.prevetmed.2009.11.001.

Rule #10: Always introduce incoming and home-produced gilts via quarantine with administration of PRRS MLV vaccination upon entry to the quarantine area

Maintaining a flow of gilt replacements into a herd is a pillar of stable production since a **sow herd with stable health performs the best**. As a result, one of the most important strategies of gilt development is a good gilt health acclimation process. This becomes especially important with high annual replacement rates in large production herds where gilts make up a relatively large proportion of the productive herd, and in herds with endemic diseases, such as PRRS. **Ensuring a well controlled PRRS immunization** and exposure of gilts during their quarantine/adaptation period **is key to protect them against field viruses** and to prepare them for the natural infection challenges they are likely to experience in endemic farms.

- **Natural immunization of gilts should be avoided** since it offers a poorly controlled immunization process and allows for the re-introduction of the wild-type PRRS virus to the herd.¹
- Modified live vaccines can replicate in the host and induce an immune response similar to that induced by mildly virulent PRRSv isolates.² Therefore, **all gilts should be quarantined and immunized two times** (3 - 4 weeks apart) with an MLV vaccination.
- The virological and clinical protection afforded by MLV vaccination is considered partial against a heterologous PRRSv strains; however, in general, **vaccinated pigs experience fewer clinical signs and a viraemia of shorter duration** compared to naïve piglets when infected with field isolates.³



¹ Rathkjen and Dall Control and eradication of porcine reproductive and respiratory syndrome virus type 2 using a modified-live type 2 vaccine in combination with a load, close, homogenise model: an area elimination study. *Acta Vet Scand* (2017) 59:4 DOI 10.1186/s13028-016-0270-z.

² Pileri and Mateu *Vet Res* (2016) Review on the transmission porcine reproductive and respiratory syndrome virus between pigs and farms and impact on vaccination 47:108 DOI 10.1186/s13567-016-0391-4.

³ Cano JP, Dee SA, Murtaugh MP, Pijoan C. Impact of a modified-live porcine reproductive and respiratory syndrome virus vaccine intervention on a population of pigs infected with a heterologous isolate. *Vaccine* 462 2007;25(22):4382–91.

Rule #10

Always introduce incoming and home-produced gilts via quarantine with administration of PRRS MLV vaccination upon entry to the quarantine area

Infection & prevention chain

The infection chain®: A systematic approach to PRRS control

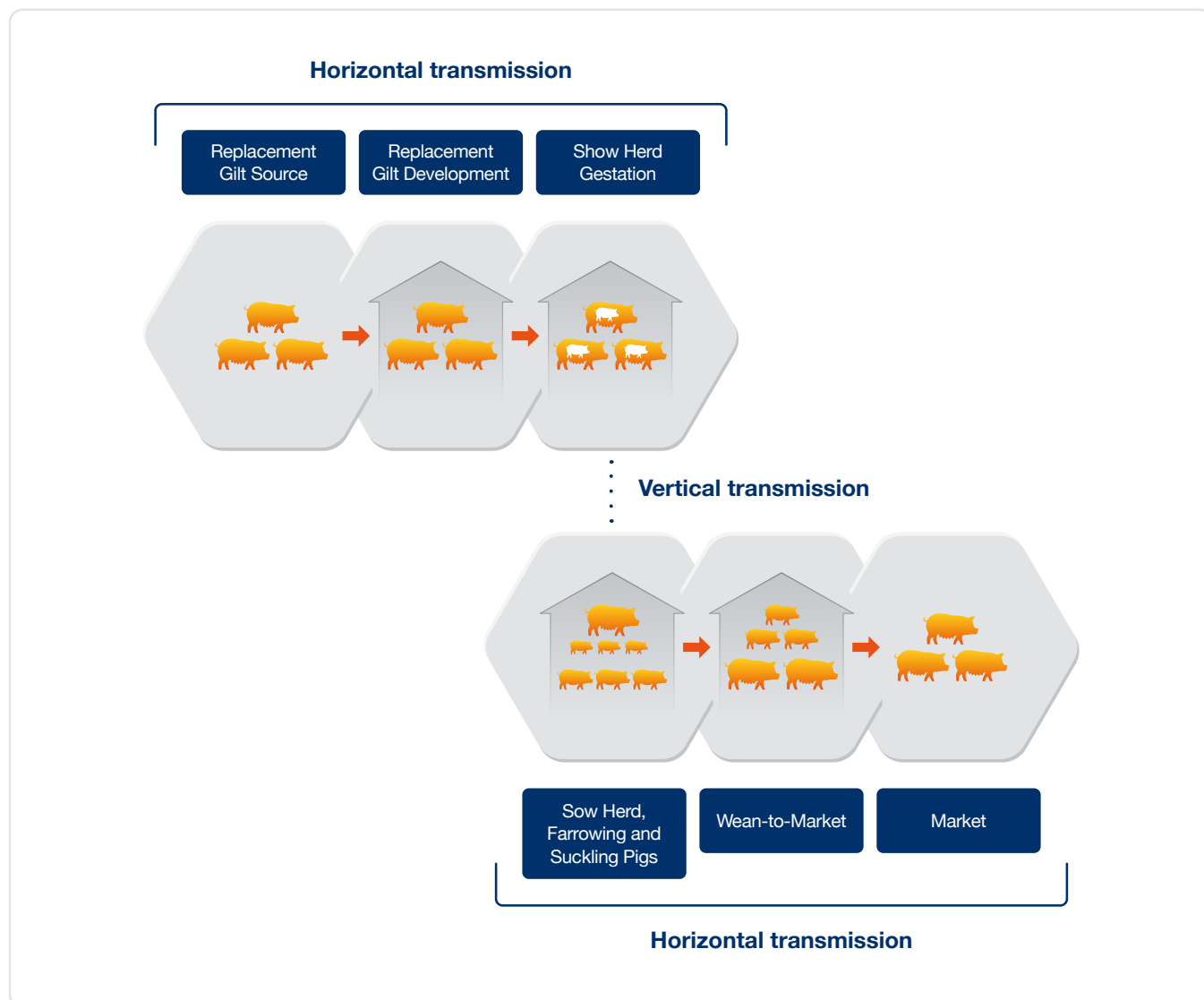
The first step in effective prrs control is identifying how and why the virus is transmitted within and among each phase of production:

- The infection chain starts with gilt development and introduction into the sow herd, and ends in grow-finish operations or reconnects with a new link to gilt production.
- Phase by phase, this tool helps you identify PRRSv status, persistence of infection, shedding and transmission patterns, as well as vertical and horizontal transmission.

By better understanding viral circulation patterns, you can create multiphase intervention strategies to target the root causes of disease. It's a holistic, logical approach to help you implement more effective prrs control and prevention programs.

The infection chain® for PRRS

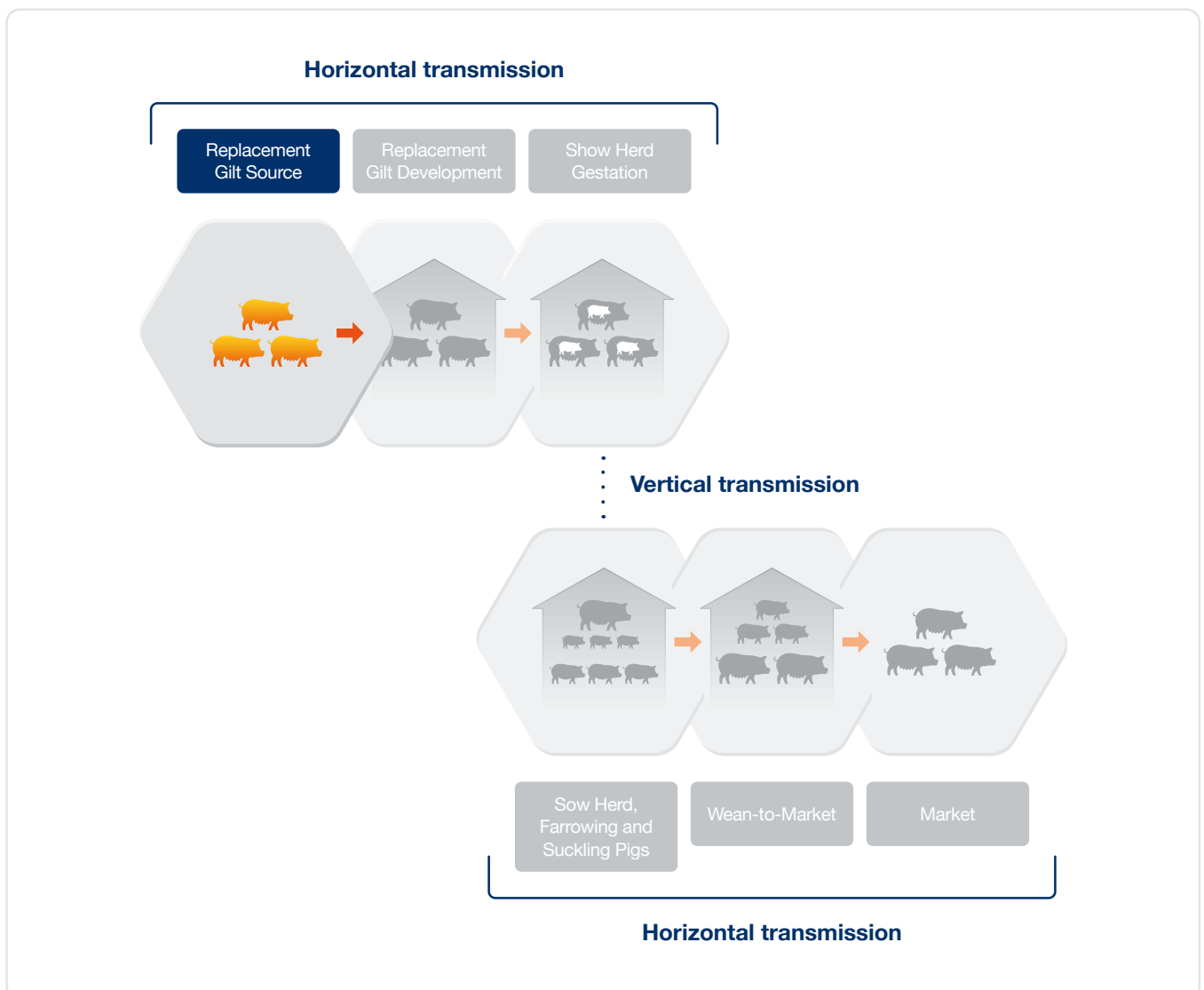
To successfully control PRRSv, it's important to first identify and understand potential causes of disease transmission, as well as opportunities for appropriate immunization at each stage of production, as part of a whole-herd approach to health management. This approach helps identify pathogen transmission patterns so diseases can be targeted at the point of infection.



Replacement Gilt Source

Goal: Purchase/procure and introduce PRRSv-negative genetic replacement gilts and semen:

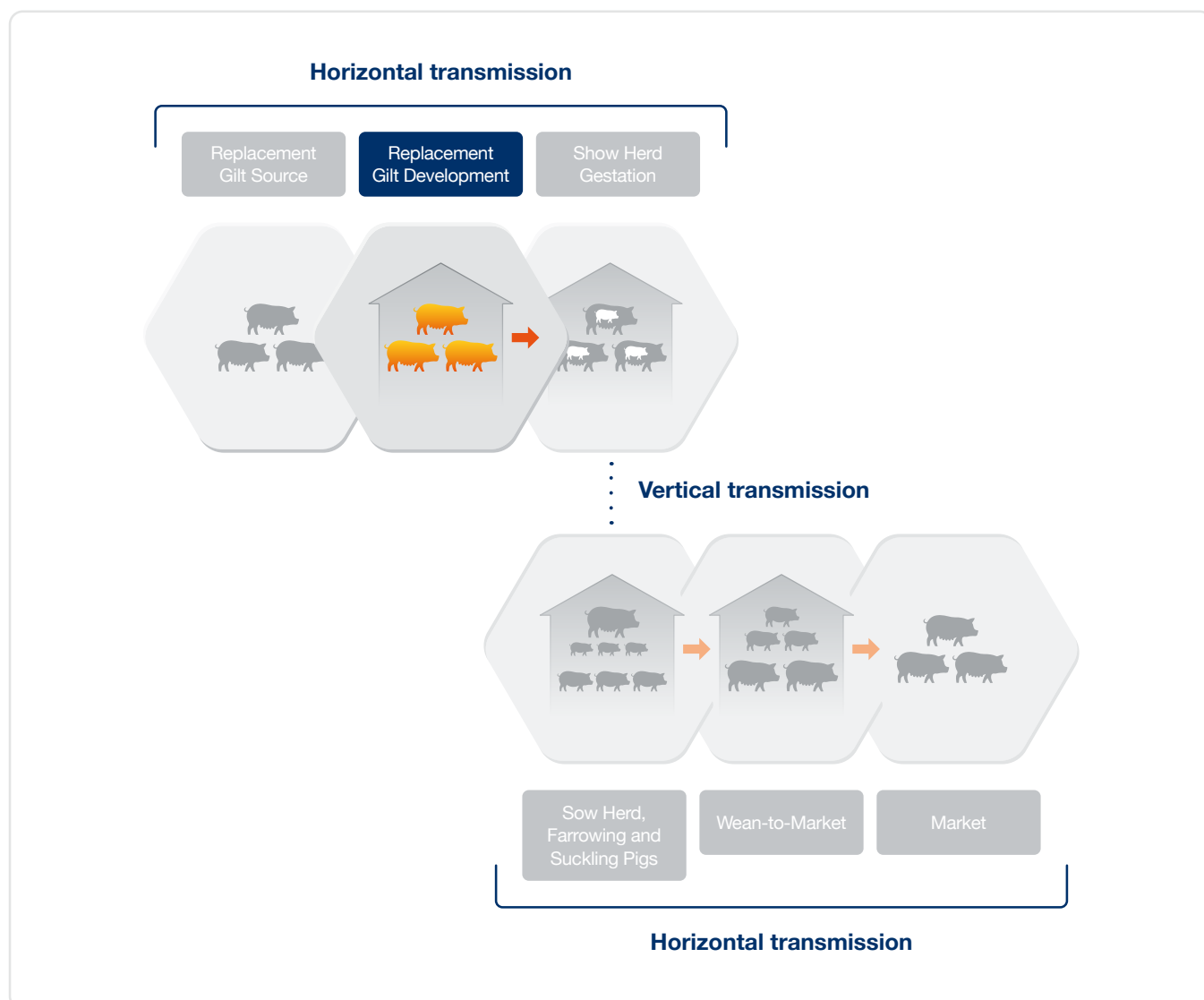
- Maintain open communication with genetic provider to know/understand current PRRSv status of gilt and semen sources at all times.
- Know and understand biosecurity protocols in place at genetic provider.
- Know and understand the PRRSv diagnostic protocol the genetic provider has in place to validate PRRSv-negative status of gilt and semen sources.



Replacement Gilt Development

Goal: Purchase or internally produce PRRSv negative gilts to use as replacement gilts. Utilize gilt acclimation/development protocols that produce an immune and non-shedding/non-infectious gilt for entry to PRRSv-positive breeding herds:

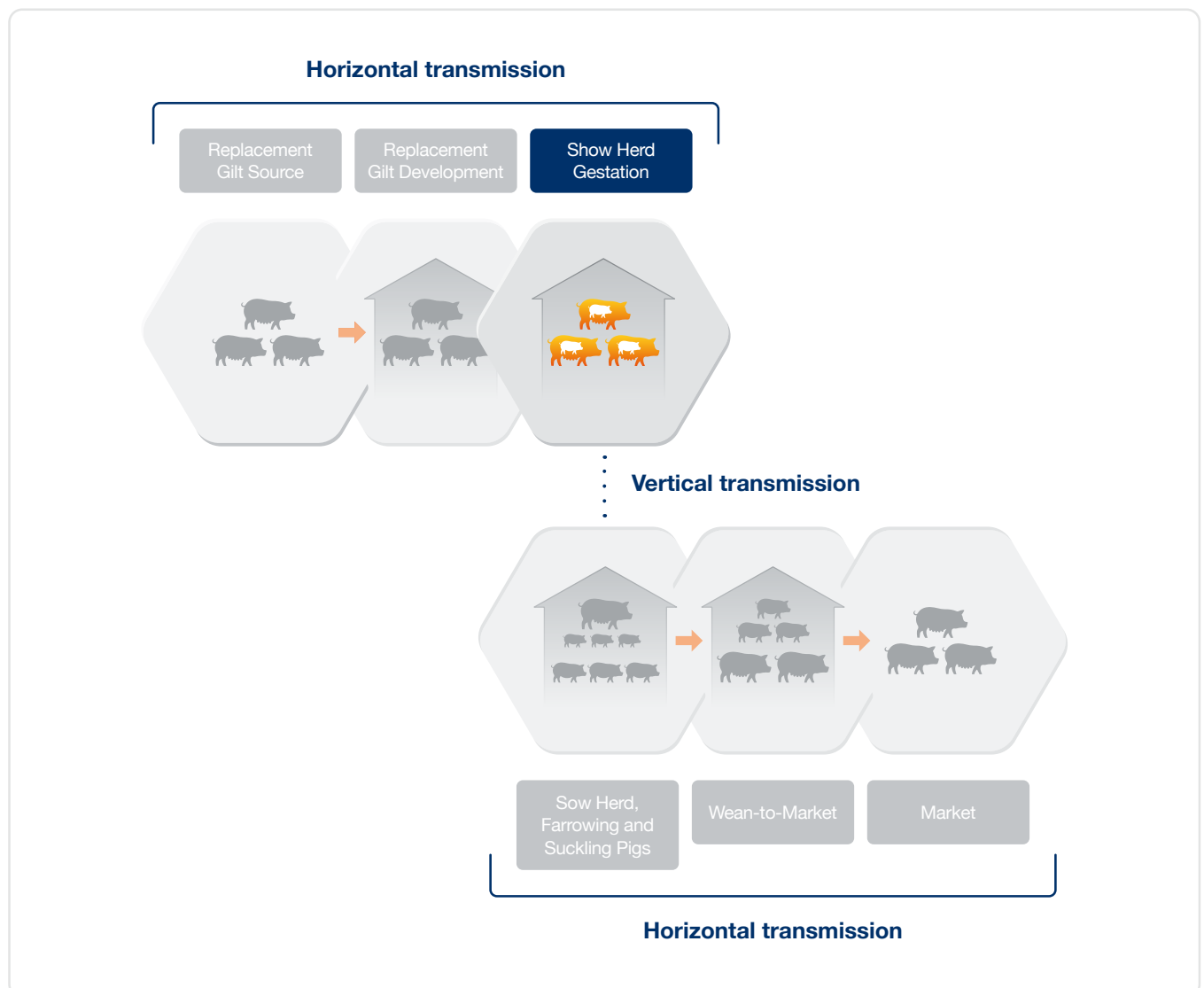
- Prior to entry into the breeding herd, vaccinate replacement gilts with Ingelvac PRRS® MLV according to the table at right.
- Validate the PRRSv-negative status at entry into the gilt development unit (GDU).
 - Serum and/or oral fluid sampling for PRRS ELISA and PRRS polymerase chain reaction (PCR) testing.
- Validate the immune and non-infectious/nonshedding status at exit from the GDU.
 - Serum and/or oral fluid sampling and PCR testing.



Sow Herd Gestation

Goal: Implement a PRRS control protocol/strategy to achieve and maintain stability with uniform PRRS immunity to mitigate/prevent the vertical transmission of virus from sow to fetus/offspring, as well as horizontal transmission from sow to sow:

- Mass-vaccinate breeding herd quarterly with Ingelvac PRRS® MLV to maintain PRRS stable status, or implement semiannual/seasonal mass vaccination protocol. Protocol dependent on specific needs/risks of the breeding herd (refer to chart at right).
 - *Maintenance of uniform-population breeding-herd immunity will reduce/eliminate resident virus circulation, and will protect and mitigate consequences of external introduction of non-resident heterologous PRRSv should it occur.*
- Diagnostically monitor PRRSv status of breeding herd/gestation herd.
 - *Refer to monitoring guidelines of the farrowing unit.*



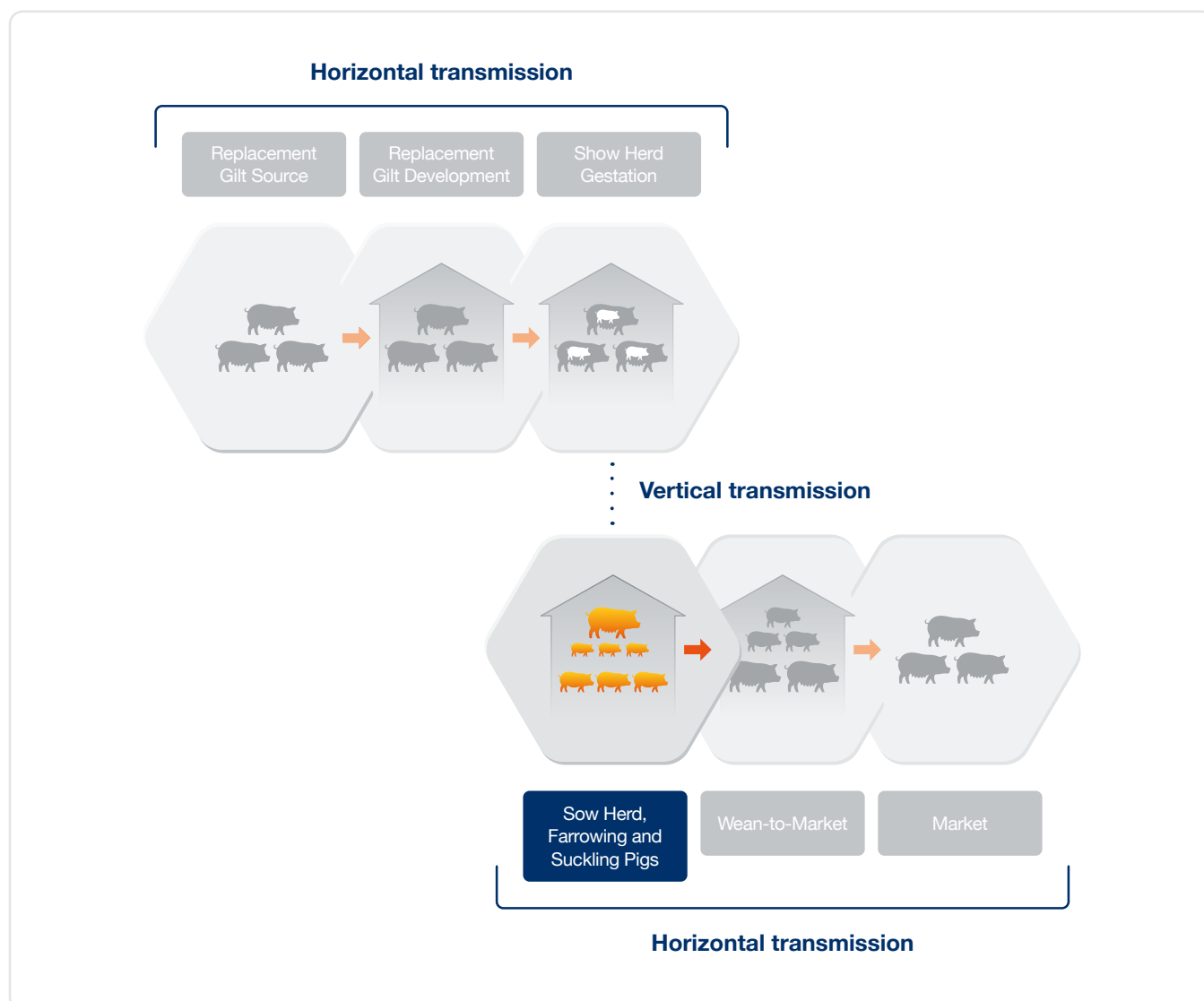
Sow Herd, Farrowing and Suckling Pigs

Goal: Implement a PRRS control protocol/strategy to achieve and maintain PRRS stable status:

- Suckling Pigs: For optimum PRRS control/protection, PRRS vaccination should occur three to four weeks prior to exposure to field virus.
 - Vaccinate with Ingelvac PRRS® MLV or 3FLEX® pre-weaning if risk of PRRSv exposure is high during the early nursery phase of production, in an effort to appropriately place vaccine three to four weeks prior to exposure/infection.
- Farrowing Room/Suckling Pigs: Implement internal biosecurity protocols to minimize/eliminate virus transmission in farrowing phase of production; e.g., McRebel-based protocols.

- Diagnostically monitor PRRSv status of breeding herd (includes farrowing and suckling pig phases of production).
- Serum-test at least 30 (in pools of five) to 60 (in pools of ten) “due-to-wean” (DTW) piglets for PRRS PCR at least monthly; sensitive to detect PRRSv at a prevalence of $\geq 5-10\%$.
 - Minimum-protocol guideline.
 - For elimination protocols, more sensitive diagnostic monitoring methods may be required.

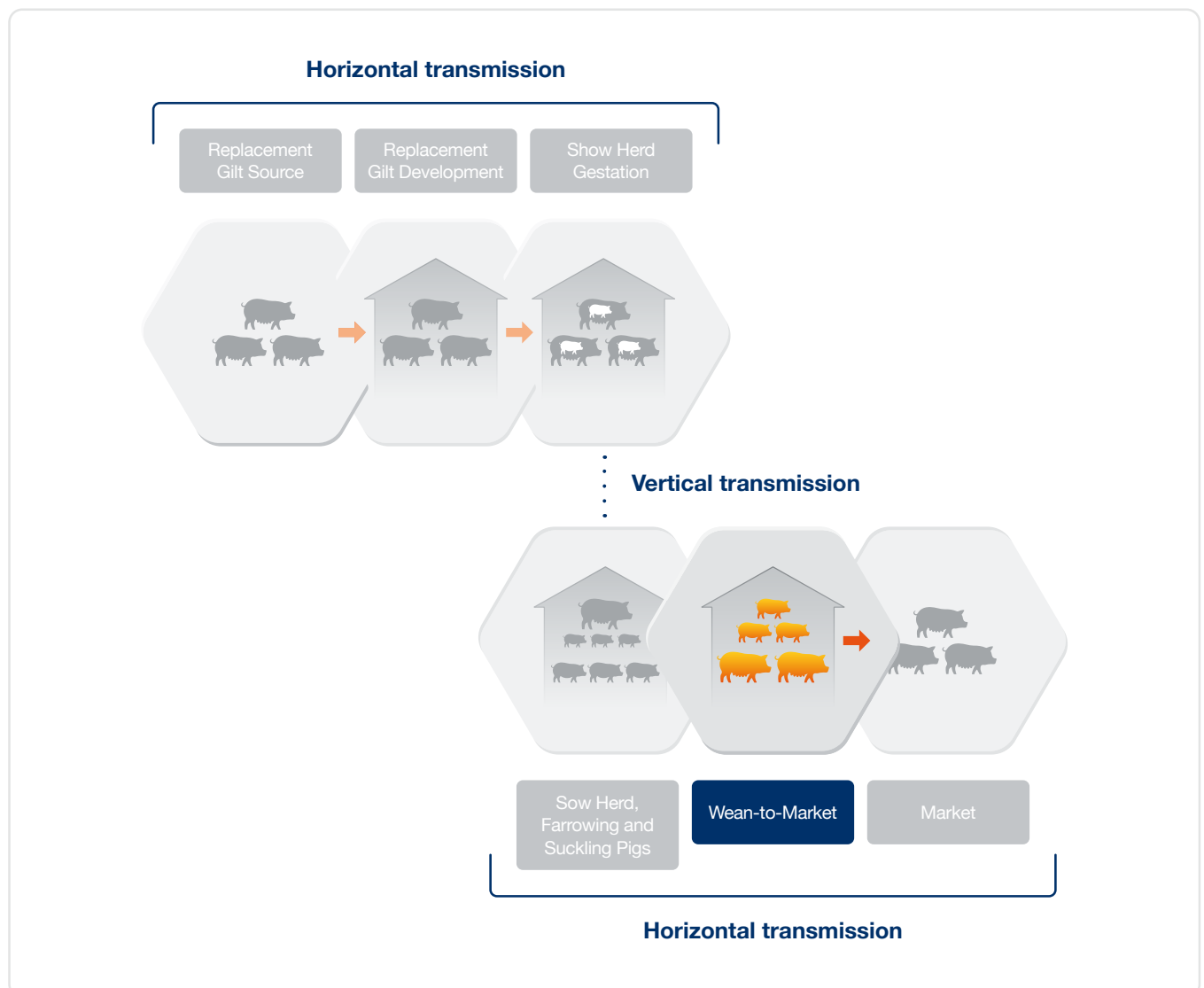
Beside Serum, other specimens such as processing fluids or tongue tip fluids can effectively monitor sow herd stability. Find out more about these specimen in the section 1.0 (page 7).



Wean-to-Market

Goal: Maximize immunity in pigs that are at risk of PRRSv exposure/infection during the wean-to-market phase of production, to mitigate the consequences of infection and improve health and performance:

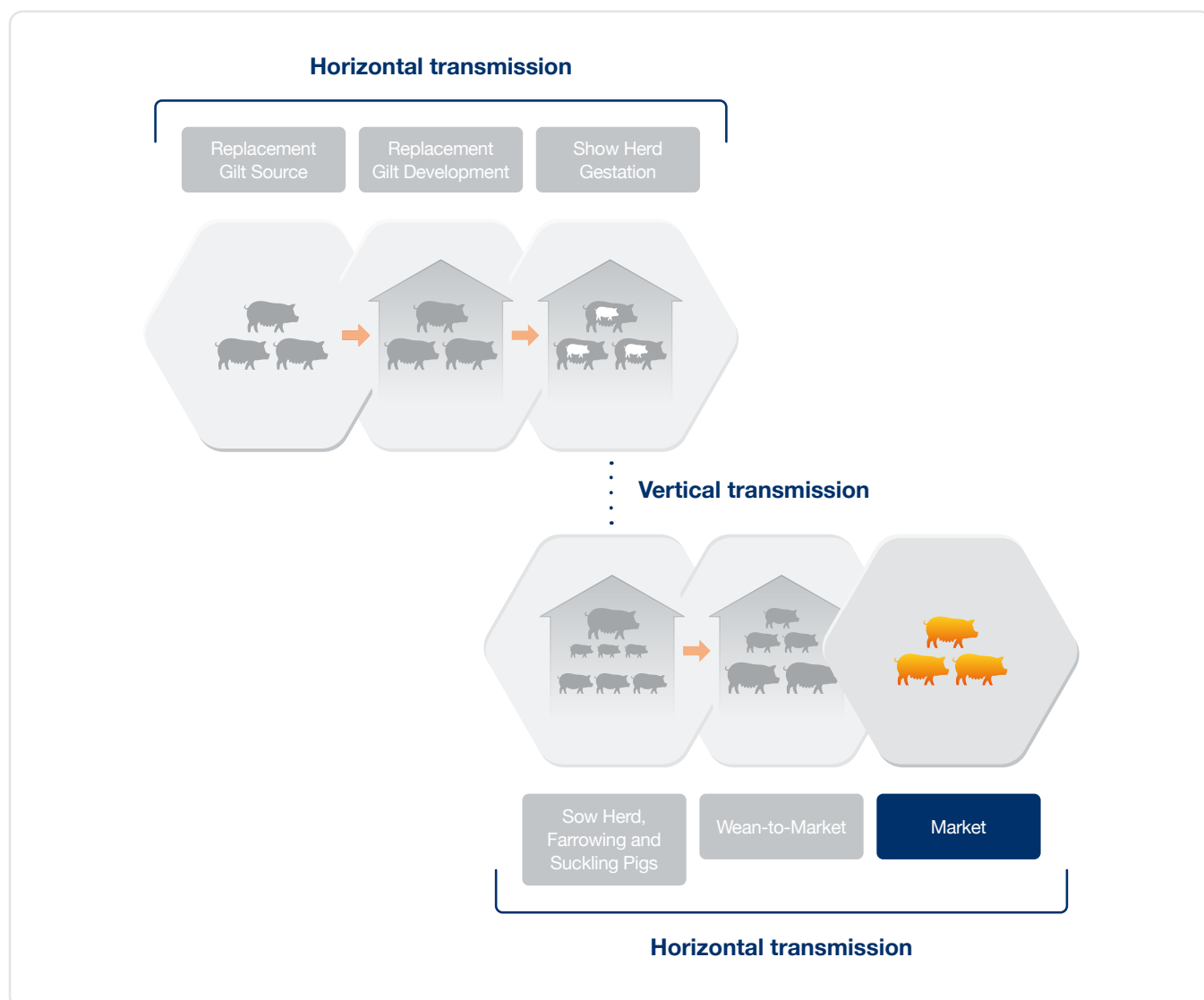
- For optimum PRRS control/protection, PRRS vaccination should occur three to four weeks prior to exposure to field virus.
 - *Vaccinate piglets with Ingelvac PRRS® MLV or 3FLEX® at or following weaning when there is risk of PRRSv infection.*
 - *Monitor performance and PRRSv status:*
 - **Performance:** Average daily gain, mortality, culls, prime marketings, etc.
 - **PRRSv status:** PRRS PCR via oral fluids at key phases of wean-to-market flow.
- Eight to 10 weeks of age (nursery exit)/12–14 weeks of age/16–18 weeks of age.



Market

Goal: Minimize/prevent exposure to virus returning to farm:

- Implement and adhere to biosecurity protocols targeted at the prevention of virus transmission from market to farm (i.e., people, fomites and transport).
- Audit external biosecurity protocols.



PRRS diagnostic Checkpoints

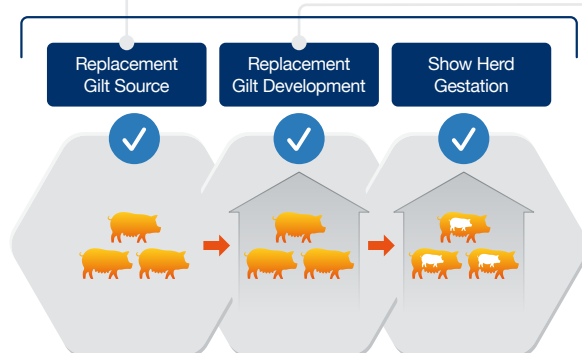
Replacement Gilt Source.

- Understand the processes your seed stock producer has in place to ensure replacement gilts are PRRSV-negative before they enter your facility. Confirm and validate the PRRSV-negative status of replacement animals with.
- PRRS ELISA and PCR testing following arrival — preferably in an isolation facility — and prior to entry into your replacement animal development and acclimation facility/phase of production.

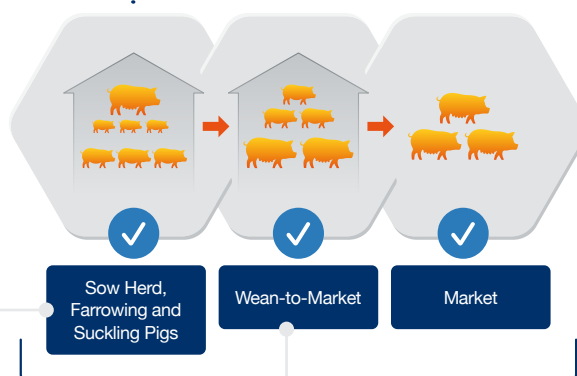
Replacement Gilt Development.

- Once you have verified that gilts are PRRSV-negative, they can enter the gilt development and acclimation phase of production.
- Gilts are vaccinated twice prior to entry to the breeding herd to introduce immune and non-infectious gilts for maintenance of uniform breeding herd immunity and stability.
- Under ideal conditions, gilts should start the acclimation process at least 12 weeks before introduction to the sow herd in a closed all in/ all out gilt flow development facility.
- At the end of the gilt development phase of production and prior to selection and entry to the breeding herd, perform PRRS ELISA, PCR and sequence testing to ensure that you are selecting and introducing immune and non-shedding/non-infectious gilts into the breeding herd.

Horizontal transmission



Vertical transmission



Gestation and farrowing/lactation phase of sow herd

- Before vaccination, collect 30 to 60 serum samples from “due-to-wean” (DTW) piglets (pools of 5 or 10) at least monthly for PCR and sequencing.
- This is a sensitive sampling method that can detect the presence of PRRSV at a prevalence level of 5–10%. For elimination protocols, more sensitive methods may be required.
- The detection and presence of PRRS-positive PCR samples is evidence of vertical and/or horizontal transmission/circulation of PRRSV in the gestation and/or lactation-farrowing phase of production.
- Evidence of PRRSV circulation may require further assessment of the stabilization protocols for gestation and farrow/lactation phases of production, which can include immunity management/vaccination protocols as well as farrowing room biosecurity protocols.

Horizontal transmission

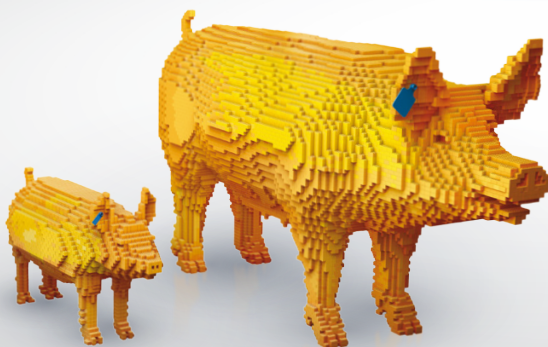
Nursery-to-finish pigs

- For nursery-to-finish pigs from stable or positive herds and flows, monitor performance and test pigs at ages 8–10 weeks, 12–14 weeks and 16–18 weeks.
- Example of testing protocol: Collect oral fluids from 1–4 ropes per 1000 pigs or air space for PCR and sequencing to assess the PRRS status of growing pig flows.



PRRS control

The power to X-protect



Discover iXt

The interactive Cross-protection tool (iXt) summarizes plenty of knowledge on PRRS on both, the sow and piglet side.

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