European PRRS Research Awards

All winning projects

Participate in the future of PRRS control!

Boehringer Ingelheim

European PRRS RESEARCH AWARD

Total number of projects funded:

36
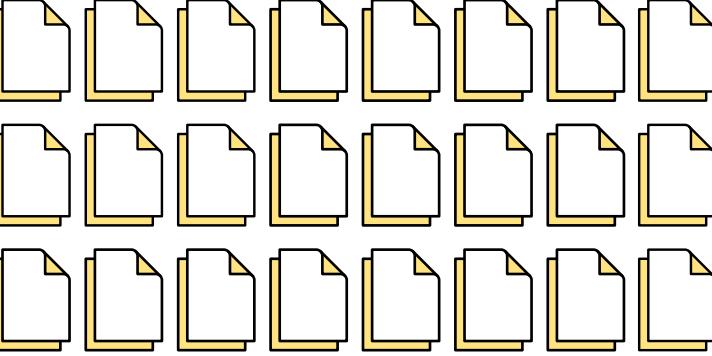
900,000€

Donated for applied reasearch on PRRS
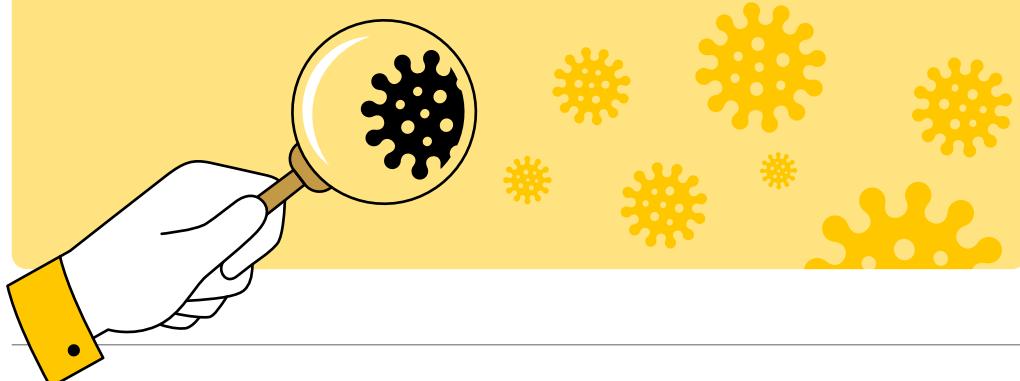
Distribution of winning projects by country

- 16
- 7
- 6
- 4
- 4
- 4
- 1
- 1

Distribution of winning projects by category

Biosecurity	Control/Management	Diagnostics	Immunity	Vaccination
5	8	10	5	8

Boehringer Ingelheim
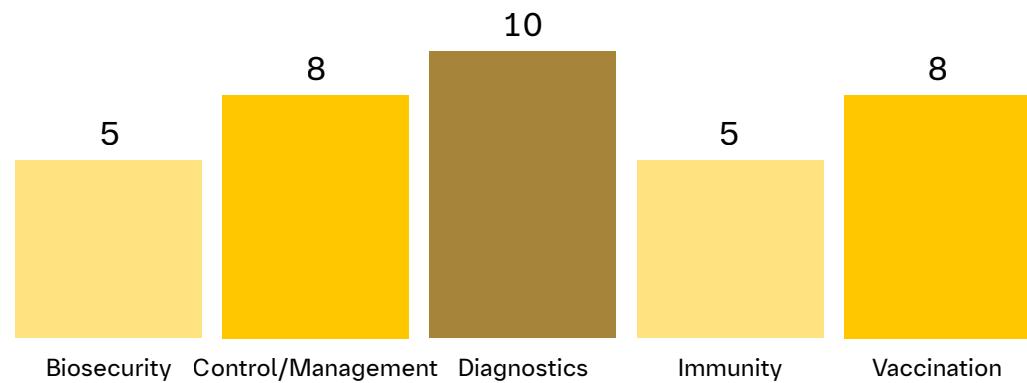
Introduction

What are the European PRRS Research Awards about?

The Boehringer Ingelheim European PRRS Research Awards are intended to help develop projects aimed at improving our knowledge of PRRS and that may have a practical application for controlling PRRS. Boehringer Ingelheim honors three research proposals with a total funding of **75,000 €** (25,000 euros each).

The independent European PRRS Research Award review board is chaired by Enric Mateu (Universidad Autonoma de Barcelona) with members from across swine practice and academia: Julia Stadler (LMU Munich), Nicolai Rosager Weber (Danish Agriculture & Food Council), Giovanbattista Danilo Guadagnini (PigVet), Carles Vilalta (IRTA-CReSA) and Joachim Seelhoff (Vet-Team Vechta). Each year the 3 most promising proposals, addressing burning PRRS related topics, get funded.

Why are they important?

In order to achieve better control of the disease, it is essential to improve our understanding of different aspects of the disease, from its epidemiology to its immunology, through the management of infected herds or monitoring methods. This knowledge has elements of basic science but, at the same time, it has to be very applied so that it can help solve practical problems that farms face. The spirit of these awards is to promote this combination of quality research with practical application, which results in relevant projects and impact for the sector. It's a win-win for all.

How to apply?

It is very easy. Every year the call is open before the summer. People interested to apply only need to download the format from *www.prrs.com* and follow the submission instructions available at the website. The proposal must contain a project description, a brief description of the originality and innovation and the practical value of the proposal. A budget and a CV of the applicant are also requested.

▶ Episode: 4

PRRS research: Panel picks latest PRRS award winners

—Prof. Enric Mateu

26 min.

"We would like to have more proposals from the field"

Professor Enric Mateu (UAB/CRESA, Barcelona, Spain) chairs the independent expert panel that judges applications for the annual European PRRS research awards sponsored by Boehringer Ingelheim. Here he outlines three winning proposals and invites more veterinary practitioners to consider applying.

Listen now:

Listen on Google Podcasts   Listen on Apple Podcasts   LISTEN ON Spotify

WATCH ON YouTube

Introduction

Detailed information for application:

Provide the following information for each project proposal (12 point type, double spaced, and a maximum of eight pages).

Outline for Proposals:

1 Complete and send the information to
olga.weber@boehringer-ingelheim.com.
Please send also a 1-page Curriculum Vitae.

2 **Project Description:**

– **Current status of problem.** Describe the significance of the problem, and summarize the current knowledge and status of the problem.

– **Related research or experience of the investigators.** Describe contributions or experience related to the proposal's topic.

– **Project objectives.** List multiple objectives separately. State the research question to be answered in each objective.

– **Procedures to achieve the objectives.** Include details of Experimental Design and Methods. Describe how the assays, procedures, and statistical tests will be done. For example, by following published procedures that are cited, or unpublished procedures that are detailed in the proposal, or by submitting samples to an established service laboratory. Briefly explain key limitations or what might go wrong, and any alternative plan to overcome the problem.

3 **Originality and innovation.** Briefly explain what is novel about the proposal.

4 **Schedule/timeline for proposed research.**

5 **Value and practical benefits of the proposed research to the swine industry.**

6 **Budget for Project:**
– Explain the budget for proposal in regards of: **Personnel, Expendables, Indirect costs, Travel, Equipment and Other.**

– If the proposal cost exceeds **25,000 €**, describe the funding available to support the total cost.

7 **Letter of recommendation (Optional).** Up to two letters of recommendation regarding the objective and importance will be accepted.

8 **Reach out to one of our PRRS Award consultants in case you would like to get additional support on your proposal.**

Introduction

Further support needed?

> **To encourage Veterinarians in the field to preapare impactful proposals we offer the expertise of external consultants supporting your project application. Please feel free to contact on of the following consultants and they will assist you to prepare your submission to the European PRRS Research Award.**

Tomasz Stadejek, Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Poland. His current research is focused on diagnostic and epidemiology of PRRSv, IAV, PCV2, PCV3 and emerging porcine parvoviruses.

Contact Tomasz Stadejek to support you with your proposal

↗ *tomasz_stadejek@sggw.edu.pl*

— *Guillermo Ramis*

— *Andrea Buzanich-Ladinig*

Guillermo Ramis is Assistant Professor at the Faculty of Veterinary Medicine in Murcia, Spain. After 13 years as practitioner in a big integration company in SE Spain he moved to the academia researching in swine health and genetics, biomarkers and viral and bacterial diseases.

Contact Guillermo Ramis to support you with your proposal

↗ *guiramis@um.es*

Andrea Buzanich-Ladinig is professor for swine medicine at the University of Veterinary Medicine Vienna, Austria. Since her PostDoc period in Canada, the research of Prof. Buzanich-Ladinig focuses on PRRSv; both, basic research and also applied research on PRRSv is done by her and her team. Applied research projects include the testing of different vaccines against PRRSv, the investigation of different diagnostic procedures including different sampling methods for the detection of PRRSv, and also the involvement in the Austrian monitoring program of PRRSv.

Contact Andrea Buzanich-Ladinig to support you with your proposal

↗ *andrea.ladinig@vetmeduni.ac.at*

Index of PRRS Award recipients

2025

Detection of PRRSv at farrowing according to sample type collected from sows and their litters over time following an outbreak
- **Elisa López Grasa.** Veterinarian, Piensos Costa S.A., Spain.

From data to action: early PRRSv outbreak detection and economic impact of early detection
- **Mafalda Pedro Mil-Homens.** Universitat Autònoma de Barcelona, Spain.

Economic burden of PRRS in Southern European pig supply chains: an evidence-based framework for decision support
- **Annalisa Scollo.** University of Turin, Italy.

2024

New diagnostic approaches to detect PRRSv in different sampling material
- **Victoria Schnitzler.** Vetmeduni Vienna, Austria.

Evaluation of the airborne transmission of two PRRSv-1 strains of different virulence levels and of an affordable filtration system to prevent this transmission
- **Olivier Bourry.** Swine Virology Immunology Unit, ANSES Ploufragan Laboratory, France.

IMPACT: Introduction or Mutations of PRRSv? – Advise for Control using advanced geneTic analyses
- **Marina Meester.** Department of Population Health Sciences, Faculty of Veterinary Medicine, Utrecht University, the Netherlands.

2023

Developing a point-of-care diagnostic procedure for PRRS virus infection based on LAMP and real-time whole genome sequencing. (POC-PRRS)
- **Marti Cortey Marques.** Departament de Sanitat i Anatomia Animals, Universitat Autònoma de Barcelona.

Assessing the risk of porcine reproductive and respiratory syndrome virus introduction in finishing herds
- **Albert Finestra Uriol.** Facultat de Veterinària, Universitat de Lleida.

Air sampling method as tool for detection and surveillance of respiratory pathogens in pig herds
- **Marylène Tignon.** Service viral enzootic, re-emerging and bee diseases Sciensano, Brussels.
- **Dominiek Maes.** Ghent University, Belgium

2022

Development of an ADKAR® change management model to wean piglets free of PRRS wild type virus; understanding the farmers objections towards a PRRS free future
- **Merel Postma.** Ghent University, Belgium
- **Diedrich Hendrickx.** Dierenartsencombinatie ZuidOost Holding BV, the Netherlands

PRRSv detection by qPCR in blood samples collected in positive stable herds following mass vaccination of sows with a MLV vaccine: A descriptive study
- **Arnaud Lebret.** Porc.Spective, France
- **Emil Lagumdzic.** University of Veterinary Medicine Vienna, Austria

Gene expression of peripheral blood mononuclear cells and CD8+ T cells from gilts after PRRSv infection.
- **Emil Lagumdzic.** University of Veterinary Medicine Vienna, Austria

2021

Tongue fluids- an alternative, practical sample material to monitor PRRSv-1?
- **Sophie Dürlinger.** Vetmeduni Vienna, Austria

First field evaluation of an innovative tool for systematic PRRSv control – including a modified Holtkamp system – on farms under western European circumstances
- **Karien Koenders.** Lintjeshof veterinary practice, the Netherlands
- **Eric van Esch.** Merefelt Livestock Diagnostics the Netherlands

Managing the PRRS positive sow herd – breeding stock introduction and biosecurity
- **Hanne Bak.** SEGES, Denmark
- **Gitte Drejer.** Danvet, Denmark

2020

PRRSv-1 recombination in swine herds: an emerging risk or a hype?
- **Erhard Van der Vries.** GD Animal Health, the Netherlands

Impact of weaning procedures on PRRSv in the nursery section
- **Pia R.** Heiselberg. HyoVet, Denmark

Biosecurity and management impact on PRRS Status and economical profit: Statistical process control after evaluation and improvement
- **Guillermo Ramis.** Universidad de Murcia, Spain

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2025

Award-winning projects:

Detection of PRRSv at farrowing according to sample type collected from sows and their litters over time following an outbreak

Elisa López Grasa. Veterinarian, Piensos Costa S.A., Spain.

From data to action: early PRRSv outbreak detection and economic impact of early detection

Mafalda Pedro Mil-Homens. Universitat Autònoma de Barcelona, Spain.

Economic burden of PRRS in Southern European pig supply chains: an evidence-based framework for decision support

Annalisa Scollo. University of Turin, Italy.

EuropeanPRRS
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2025

Elisa López Grasa.
Veterinarian, Piensos Costa S.A., Spain.

Detection of PRRSv at farrowing according to sample type collected from sows and their litters over time following an outbreak

Current problem

Clinical suspicion of PRRS should be considered when there is a sudden increase in abortion rates, a significant reduction in the number of live-born piglets, and/or a rise in piglet losses observed on sow farms.

Traditionally, the recommended target population for determining the status of breeding herds has been weaned piglets. However, this monitoring system does not allow us to distinguish between vertical transmission (from sows to newborn piglets) and horizontal transmission among suckling piglets.

The presence of subpopulations after an outbreak caused by a new strain could maintain viral circulation at a low level. Some aspects of PRRSV infection dynamics in the sow population remain unclear, such as the time it takes to reach low prevalence in sows.

Study objective

Primary objective
Determine the most effective sample type (sow TOSCs, sow serum, stillborn TTF and serum, live-born serum) for detecting PRRSV at farrowing over time following an outbreak.

Secondary objective
Compare sample types (sow TOSCs, sow serum, stillborns TTF, stillborn serum, live-born serum) regarding PRRSV detection, cycle threshold (Ct) values, and parity.

Results

The study is currently being conducted. Results will be published in scientific literature and upcoming events.

2025

Mafalda Pedro Mil-Homens.
Universitat Autònoma de Barcelona,
Spain.

From data to action: early PRRSv outbreak detection and economic impact of early detection

Current problem

Surveillance methods for PRRS have evolved from traditional serum-based diagnostics to more practical sample types like oral fluids, processing fluids, and tongue tips. Additionally, there's a growing shift toward using production-based data (e.g., farrowing rates, mortality, feed intake) for early outbreak detection — an approach known as syndromic surveillance.

Despite its potential, syndromic surveillance is underutilized in Spanish swine herds. Early detection through monitoring production anomalies can prompt faster interventions, reduce disease spread, and mitigate economic losses. The proposal aims to enhance this approach using multivariate time series analysis and change-point detection, which are still underexplored in swine health management.

Study objective

1. Early Detection of PRRSv Outbreaks
Apply multivariate time series analysis combined with change-point detection to identify early signs of PRRSv outbreaks in swine breeding herds. This will be done by analyzing production data (e.g., abortions, mortality, sow deaths, litters weaned) to detect unusual patterns that may signal the onset of an outbreak.

2. Assessing the Impact of Early Detection
After validating the detection models, the project will evaluate how early detection affects:

- Disease spread to other farms

- Production losses

This analysis will quantify the benefits of early intervention and support decision-making in swine health management.

Results

The study is currently conducted. Results will be published in scientific literature and upcoming events.

Annalisa Scollo.
University of Turin, Italy.

Economic burden of PRRS in Southern European pig supply chains: an evidence-based framework for decision support

Current problem

Porcine reproductive and respiratory syndrome (PRRS) remains one of the most persistent and economically damaging diseases in global pig production. Its impact includes:

- **Direct losses:** reproductive failure, mortality, reduced growth.
- **Indirect costs:** longer time to market, lower feed efficiency, increased antimicrobial use.

Although control measures like vaccination and biosecurity exist, their cost-effectiveness is hard to evaluate due to limited real-world economic data—especially in Southern Europe, where:

- High farm density and climate conditions favour viral spread.
- More virulent strains exacerbate losses.
- Existing economic models (mostly from North America or Northern Europe) lack applicability.

Previous European studies (e.g., Nathues *et al.*, Renken *et al.*) provided estimates using simulations or expert opinion but lacked validation with real-time farm data.

Study objective

The project aims to develop and validate a data-driven framework to estimate the economic impact of PRRS in Southern European pig production systems, using real-world data and advanced analytics.

Objective 1: Data Integration

- Build an automated infrastructure to consolidate diverse farm data (e.g., productivity, diagnostics, treatments, market data).

- Harmonize this information into a master dataset suitable for economic modeling.

Objective 2: Economic Modelling

- Develop a full-budget economic model tailored to Southern European conditions.
- Quantify financial losses at batch and herd levels due to PRRS outbreaks.

Objective 3: Causal Inference

- Apply statistical methods to isolate the true impact of PRRS from confounding factors (e.g., seasonality, coinfections).
- Determine how much of the observed performance loss is directly attributable to PRRS.

Objective 4: Scenario Analysis

- Combine causal inference and economic modeling to simulate outbreak vs. non-outbreak scenarios.
- Translate results into actionable financial insights for producers and managers.

Objective 5: Field Validation

- Test the model in commercial farm settings.
- Compare predicted vs. observed losses and assess usability as a decision-support tool for veterinarians and farmers.

Results

The study is currently conducted. Results will be published in scientific literature and upcoming events.

2024

Award-winning projects:

New diagnostic approaches to detect PRRSv in different sampling material

Schnitzler. Vetmeduni Vienna, Austria.

PRRS'R: Evaluation of the airborne transmission of two PRRSv-1 strains of different virulence levels and of an affordable filtration system to prevent this transmission

Olivier Bourry. Swine Virology Immunology Unit, ANSES Ploufragan Laboratory, France.

IMPACT: Introduction or Mutations of PRRSv? – Advise for Control using advanced geneTic analyses

Marina Meester. Department of Population Health Sciences, Faculty of Veterinary Medicine, Utrecht University, the Netherlands.

EuropeanPRRS RESEARCH AWARD

2024

Victoria Schnitzler.
Vetmeduni Vienna, Austria.

New diagnostic approaches to detect PRRSv in different sampling material

Current problem

PRRSv causes significant economic losses in pig production, with diagnostic challenges due to low viral loads, particularly in endemic farms.

Effective control relies on monitoring sow herds using standardized sampling methods like serum, processing fluids, and oral fluids. Additionally, new alternative samples, such as tongue fluids from stillborn or dead suckling piglets, are being explored. Ensuring the stability of viral RNA during transport is crucial for accurate PCR and sequencing results, highlighting the need to evaluate RNA stabilizers across different sample types to improve diagnostic methods.

Study objective

The study aims to evaluate different transport media for stabilizing PRRSv RNA in various sampling materials to enhance viral RNA detection via PCR and facilitate genome sequencing. Various samples, including serum from weaned piglets and oral fluids, will be analyzed with and without stabilizers. The study will compare viral concentrations and monitor virus detection after an outbreak in different sample materials and under varying storage conditions.

Additionally, sequencing methods such as Sanger sequencing, Next-Generation Sequencing (NGS), and nanopore sequencing will be examined to provide recommendations for effective PRRSv monitoring.

Results

The study is currently conducted. Results will be published in scientific literature, and upcoming events.

Olivier Bourry.
Swine Virology Immunology Unit,
ANSES Ploufragan Laboratory,
France.

Evaluation of the airborne transmission of two PRRSv-1 strains of different virulence levels and of an affordable filtration system to prevent this transmission

Current problem

PRRSv can be transmitted within or between farms through various routes.

While the airborne transmission of PRRSv-2 strains has been confirmed, the airborne transmission of PRRSv-1 strains remains hypothetical. This gap in understanding poses a challenge for developing effective control strategies to prevent the spread of PRRSv-1 in pig populations.

Study objective

This project aims to investigate the airborne transmission of two PRRSv-1 strains with different levels of virulence (low and high pathogenic strains) under controlled experimental conditions.

Should airborne transmission be confirmed for at least one of these strains, the next phase of the study will focus on evaluating the effectiveness of an affordable and simple filtration system to prevent this mode of transmission.

Results

The study is currently conducted. Results will be published in scientific literature, and upcoming events.

2024

Marina Meester.
Department of Population Health
Sciences, Faculty of Veterinary
Medicine, Utrecht University,
the Netherlands.

IMPACT: Introduction or Mutations of PRRSv? – Advise for Control using advanced geneTic analyses

Current problem

Controlling PRRSv is complicated by the virus's high genetic variation. The dominant strain of PRRSv in a farm can shift due to mutations, recombination with other field or vaccine-like strains, or the introduction of a completely new strain. This genetic complexity makes interpreting PRRSv sequence data challenging, yet farmers increasingly rely on sequencing to track PRRSv dynamics and adjust their management practices.

Study objective

The IMPACT project aims to utilize advanced genetic methods to differentiate between mutations of circulating PRRSv strains and newly introduced or vaccine-like strains.

By analyzing the virus's evolutionary rate, the study will also determine when new strains were introduced into farms and correlate these events with changes in biosecurity measures and vaccination protocols, offering more precise guidance for PRRSv control.

Results

The study is currently conducted. Results will be published in scientific literature, and upcoming events.

2023

Award-winning projects:

➡ **Developing a point-of-care diagnostic procedure for PRRS virus infection based on LAMP and real-time whole genome sequencing. (POC-PRRS)**

Marti Cortey Marques. Departament de Sanitat i Anatomia Animals, Universitat Autònoma de Barcelona.

➡ **Assessing the risk of porcine reproductive and respiratory syndrome virus introduction in finishing herds**

Albert Finestra Uriol. Facultat de Veterinària, Universitat de Lleida.

➡ **Air sampling method as tool for detection and surveillance of respiratory pathogens in pig herds**

Marylène Tignon. Service viral enzootic, re-emerging and bee diseases Sciensano, Brussels.

Marti Cortey Marques.
Departament de Sanitat i Anatomia Animals, Universitat Autònoma de Barcelona.

Developing a point-of-care diagnostic procedure for PRRS virus infection based on LAMP and real-time whole genome sequencing. (POC-PRRS)

Current problem

Time is a crucial factor in diagnostics. Rapid results are essential for effective decision-making in the field. Point-of-care diagnostics, which allow testing to be performed directly on-site, help reduce the time needed for results. However, such testing requires simplified diagnostic methods, such as loop-mediated isothermal amplification (LAMP) and real-time next-generation sequencing (NGS).

Study objective

The primary goal of the POC-PRRS project is to develop a point-of-care methodology for diagnosing and character-izing PRRSv-1, to be applied on-site using portable tools and devices, with results generated within approximately 24 hours. Once established, the second objective is to validate this methodology using field samples collected for monitoring and diagnostic purposes.

Results

The study is currently conducted. Results will be pub-lished in scientific literature, and upcoming events.

Albert Finestra Uriol.
Facultat de Veterinària, Universitat de Lleida.

Assessing the risk of porcine reproductive and respiratory syndrome virus introduction in finishing herds

Current problem

PRRS is one of the most economically damaging diseases in pig production, also impacting animal welfare and the morale of both farmers and veterinarians. Despite efforts, the disease, which has plagued the industry since 1991, continues to elude a consistent solution.

Study objective

This study focuses on a critical concern: understanding how PRRSv enters farms. It aims to investigate the potential entry routes into finishing herds that introduce negative piglets into areas with high and low pig density, helping to identify and address vulnerabilities in farm biosecurity.

Results

The study is currently conducted. Results will be published in scientific literature, and upcoming events.

Marylène Tignon.
Service viral enzootic, re-emerging and bee diseases Sciensano, Brussels.

Dominiek Maes.
Ghent University, Belgium

Air sampling method as tool for detection and surveillance of respiratory pathogens in pig herds

Current problem

Infections from respiratory pathogens pose significant health and economic risks in pig-producing countries. Pneumonia constitutes the leading cause of death in pigs, with financial losses linked to reduced performance, carcass quality, and increased treatment costs. Respiratory diseases also raise public health concerns due to their role in antimicrobial use, antibiotic resistance, and zoonotic risks, highlighting their relevance in the "ONE HEALTH" approach.

The porcine respiratory disease complex (PRDC) involves multiple pathogens, including PRRSv, and is often exacerbated by environmental and management factors. Current strategies have focused narrowly on individual pathogens, lacking comprehensive surveillance and control measures. Existing diagnostic methods are labor-intensive and invasive, necessitating more efficient, non-invasive techniques.

Air sampling methods could improve monitoring, reduce costs, and enable quicker surveillance of emerging pathogens, ultimately supporting better animal health and welfare management.

Study objective

The study investigates the potential of bioaerosols for the surveillance and monitoring of major respiratory pathogens such as PRRSv, swine influenza, and M. hyopneumoniae in the air of pig barns, as a replacement or complementary tool for traditional individual and invasive sampling methods.

Summary

The study is currently conducted. Results will be published in scientific literature, and upcoming events.

2022

Award-winning projects:

→ **Development of an ADKAR® change management model to wean piglets free of PRRS wild type virus; understanding the farmers objections towards a PRRS free future**

Merel Postma. Ghent University, Belgium
Diedrich Hendrickx. Dierenartsencombinatie ZuidOost Holding BV, the Netherlands

→ **PRRSv detection by qPCR in blood samples collected in positive stable herds following mass vaccination of sows with a MLV vaccine: A descriptive study**

Arnaud Lebret. Porc.Spective, France

→ **Gene expression profiling of peripheral blood mononuclear cells and CD8+ T cells from gilts after PRRSv infection.**

Emil Lagumdzic. University of Veterinary Medicine Vienna, Austria

European**PRRS**
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AWARD

Merel Postma.
Ghent University, Belgium

Diedrich Hendrickx.
Dierenartsencombinatie ZuidOost
Holding BV, the Netherlands

Development of an ADKAR® change management model to wean piglets free of PRRS wild type virus; understanding the farmers objections towards a PRRS free future

Current problem

PRRS control was proven to be highly relevant for sustainable pig production. The success of any control program is highly dependent on willingness of farm workers to implement the program. Farm personal is not always motivated to follow a control plan that might initially result in additional workload.

Study objective

To be able to use Hiatt's ADKAR®-model of change for farm based consultancy on PRRS in a way that the veterinary practitioner can use the model to objectively determine what are the constraints in a farm to improve the PRRS-status.

Summary

Results indicated that very few farmers were mentally prepared to eliminate PRRSv: For 57% of the farmers, perception and/or motivation were expected to limit successful change. For 70% of the farmers, knowledge and for 52% of the farmers, a lack of ability were the limiting factor. The ADKAR model proved useful for identifying the key elements that prevent successful behavioral change.

Practical benefit

The higher the number of farmers that implement a well thought over and tailor made sustainable PRRS control plan in their farms, the sooner the Netherlands will be close to PRRS wild type free husbandry.

Should we be able to facilitate this goal for individual farms by means of a practical and motivating system the more successful we will be. If the consulting veterinarian, who in the Netherlands is the first contact for animal diseases and above all is the veterinary responsible person in all certifying programs, is not capable of convincing the farmer and employees, then who is?

Further reading

↗ *The ADKAR® change management model.*

↗ *The human factor: Are we ready to eradicate PRRS?*

Arnaud Lebret.
Porc.Spective, France

PRRSv detection by qPCR in blood samples collected in positive stable herds following mass vaccination of sows with a MLV vaccine: A descriptive study

Current problem

Detecting PRRSv in due to wean piglets after sow mass vaccination raises questions whether the detected virus is a vaccine or a wild type virus. Few reports are available in peer-reviewed journals regarding the frequency of detection of vaccine virus in vaccinated sow herds with a MLV even if, practically, it is a recurrent concern for practitioner.

Study objective

The objective of this project is to evaluate the frequency of detection of vaccine strains in due-to-wean piglets after sow mass vaccination with two different vaccines. Porcilis® PRRS and UNISTRAIN® PRRS have been chosen for this trial due to the fact that the same trial design has previously been studied with ReproCyc® PRRS EU (Lebret *et al.* 2022, publication in progress). This study will help practitioners to precise their diagnostics strategy depending on the use of each vaccine and will complete previous study published for ReproCyc® PRRS EU.

Summary

The study is currently conducted. Results will be published at the ESPHM 2025.

Practical benefit

This study will help practitioners to precise their diagnostics strategy depending on the use of each vaccine and will complete previous study published for ReproCyc® PRRS EU.

Further reading

In a previous study *Lebret et al., 2021* analyzed blood and processing fluid samples from piglets in a PRRS sable 100 sow farm. Four consecutive batches were tested, born after a booster sow mass MLV vaccination with ReproCyc®. No PRRSv by qPCR could be detected in piglets from vaccinated sows.

↗ *PRRSv detection by qPCR in processing fluids and serum samples collected in a positive stable breeding herd following mass vaccination of sows with a modified live vaccine.*

↗ *PRRSv Detection by qPCR on Serum Samples Collected in Due-to-Wean Piglets in Five Positive Stable Breeding Herds Following a Sow Mass Vaccination with a Modified Live Vaccine: A Descriptive Study.*

2022

Emil Lagumdzic.
University of Veterinary Medicine
Vienna, Austria

Gene expression of peripheral blood mononuclear cells and CD8+ T cells from gilts after PRRSv infection.

Current problem

Alteration of the host immune response by PRRSv is associated with the increased susceptibility to secondary viral and bacterial infections resulting in more serious and chronic disease. However, the expression profiles underlying innate and adaptive immune responses to PRRSv infection are yet to be further elucidated.

Study objective

We provided extensive transcriptomics data explaining gene signatures of the immune response of PBMCs and CD8+ T cells after PRRSv infection. Additionally, our study provides potential biomarker targets useful for vaccine and therapeutics development.

Summary

We identified the highest number of differentially expressed genes in PBMCs and CD8+ T cells at 7 dpi and 21 dpi, respectively. Gene signature of PBMCs from infected animals was characterized by strong innate response at 7 dpi which continued at 14 dpi and 21 dpi but in addition involved adaptive immunity. Conversely, gene expression pattern of CD8+ T cells revealed a strong adaptive immune response to PRRSv which induced the generation of highly differentiated CD8+ T cells from 14 dpi. Moreover, the hallmarks of the CD8+ T-cell response to PRRSv were the upregulation of effector and cytolytic genes (PRF1, GZMA, GZMB, GZMK, KLRK1, KLRD1, FASL, NKG7).

Practical benefit

A total of 64 samples were included in the study. PBMCs and MACS-sorted CD8+ T cells were derived from four PRRSv-infected gilts as well as from four non-infected gilts. Blood was collected at four different time points, starting at day 0 just prior to experimental infection of the infection group, followed by blood sampling at days 7, 14 and approximately 21 (termination day, 21± 2) after infection. To gain deeper insight into the pathogenesis of PRRS we examined temporal gene expression patterns from 0 dpi to 21 dpi. Consequently, the present study includes time-series clustering analysis, protein-protein interaction (PPI) networks, extensive gene ontology (GO) enrichment and pathway analysis, to define the innate and adaptive immunity against PRRSv more accurately.

Further reading

↗ *Porcine reproductive and respiratory syndrome (PRRS): an immune dysregulatory pandemic.*

Emil Lagumdzic.
University of Veterinary Medicine
Vienna, Austria
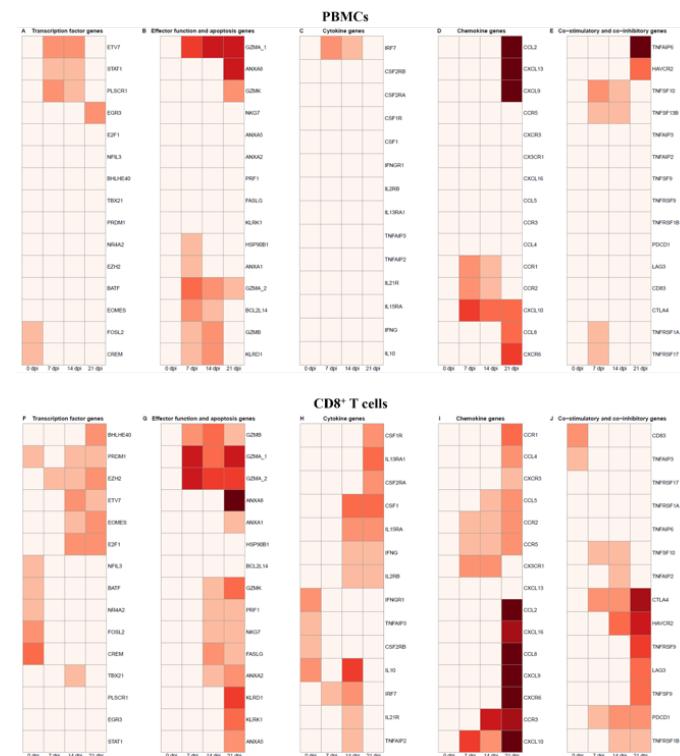
Figure 1. Transcription profiles of PBMCs and CD8+ T cells from 4 infected animals at 4 time points.

Figure 2. Temporal clustering of genes in PBMC and CD8+ T cells from PRRSv-infected animals

Discussion and Conclusion

These results elucidated the tight transcriptional regulation of the innate and the adaptive immune response to PRRSv. Compared to PBMCs, CD8+ T cells revealed a much higher number of possible biomarkers that could discriminate non-infected and PRRSv-infected gilts from 7 dpi to dpi 21.

2021

Award-winning projects:

Tongue fluids- an alternative, practical sample material to monitor PRRSv-1?

Sophie Dürlinger. Vetmeduni Vienna, Austria

First field evaluation of an innovative tool for systematic PRRSv control – including a modified Holtkamp system – on farms under western European circumstances

Karien Koenders. Lintjeshof veterinary practice, the Netherlands
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Managing the PRRS positive sow herd – breeding stock introduction and biosecurity

Hanne Bak. SEGES, Denmark
Gitte Drejer. Danvet, Denmark

EuropeanPRRS
RESEARCH
AWARD

2021

Sophie Dürlinger.
Vetmeduni Vienna, Austria

Tongue fluids- an alternative, practical sample material to monitor PRRSv-1?

Current problem

Currently, processing fluids are often used for PRRSv monitoring of a sow herd, since they are a practical, time- and cost- efficient aggregated sample material. However, the prohibition of routine tail docking in the European Union is one reason why we are still looking for new, innovative but also time- and cost-efficient PRRSv monitoring methods. Apart from that, surgical castration without anaesthesia is already banned in many European countries. Some countries are mainly using alternatives such as boar fattening or immunocastration instead of the surgical castration of male piglets. As a result, testicles of male piglets can no longer be used as material for PRRSv monitoring in some European countries. For this reason, the present study investigated whether tongue fluids are a suitable sample material for detection of PRRSv-1 by means of RT-qPCR.

Study objective

Objectives of the present project were:

1 To evaluate under experimental conditions whether foetal tongue fluids are a suitable sample material for detection of PRRSv-1 by means of RT-qPCR.

2 To find out whether tongue fluids of stillborn piglets represent a suitable sample material in the field to monitor PRRSv-1 positive breeding farms after an acute PRRSv-1 outbreak.

Study

1 **Study under experimental conditions:**

Experimentally infected (PRRSv-1 AUT15-33) pregnant gilts (day 85 of gestation) were euthanized between gestation day 104 and 110 and a detailed necropsy and sampling of the gilts and foetuses was performed. Foetal thymus and foetal serum were examined for PRRSv-specific genome fragments using RT-qPCR (Table 1). Furthermore, processing fluids pooled per litter, fluids from individual tongues (n=88) of each foetus from five infected gilts and fluids from tongue pieces pooled by litter (n=26) were examined by RT-qPCR (Table 1).

2 **Field study:**

Two farms were monitored over several farrowing groups following an acute PRRS outbreak. Litter-wise pooled tongue tissue samples from stillborn piglets and piglets that died in the first days of life were collected. Additionally, processing fluids were gathered from all piglets and pooled per litter. Furthermore, serum samples from two piglets per litter at three weeks of life were examined in pools of four to six piglets, and oral fluid samples from piglets after weaning (5th week of life) were examined for PRRSv using RT-qPCR.

2021

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Tongue fluids- an alternative, practical sample material to monitor PRRSv-1?

Results

gilt		L1	L2	L3	L4	L5	L6	L7	L8	L9	L10	L11	L12	L13		R13	R12	R11	R10	R9	R8	R7	R6	R5	R4	R3	R2	R1	gilt
1	foetal preservation status	AUT	AUT	MEC1	MEC1	MEC1	MEC2	MEC2	MEC1	MEC1	VIA									MEC1	VIA	MEC1	MEC1	MEC1	VIA	MEC2	DEC	MEC1	1
2		VIA	VIA	VIA	VIA	VIA	VIA	VIA	MEC1	VIA	MEC1	VIA	VIA	VIA		VIA	VIA	VIA		MEC2	MEC2	MEC1	AUT	VIA	MEC1	VIA	VIA	VIA	2
3		VIA	VIA	VIA	MEC1	VIA	VIA	VIA	VIA	VIA	VIA	VIA	VIA	MEC1				MEC2	MEC2	VIA	VIA	VIA	VIA	VIA	VIA	VIA	VIA	DEC	3
4		DEC	VIA	VIA	AUT	MEC1	VIA	MEC1	VIA	MEC1											AUT	VIA	MEC1	VIA	VIA	VIA	MEC1	AUT	4
5		VIA	VIA	VIA	VIA	VIA	VIA	VIA	VIA	VIA										MEC1	VIA	VIA	MEC1	VIA	VIA	VIA	VIA	MEC1	5
1	viral load (log₁₀ GE/mL) foetal serum	(-)	(-)	7.86	8.89	8.93	8.63	8.61	8.72	7.99	(*)									8.26	8.13	7.95	8.37	8.06	(*)	8.15	(-)	7.67	1
2		(*)	(*)	(*)	(*)	(*)	(*)	(*)	8.18	7.56	8.15	(*)	(*)	(*)			(*)	4.96	(*)	8.73	8.71	7.99	(-)	6.02	(*)	(*)	7.77	(*)	2
3		(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	5.23	(*)	(*)						8.59	8.67	(*)						5.35	3
4		(-)	(*)	8.63	(-)	7.84	(*)	8.45	(*)	8.59										(-)	8.21	8.60	(*)	(*)	(*)	8.26	(-)		4
5		(*)	(*)	(*)	(*)	(*)	(-)	(*)	(*)	(*)	(*)									3.84	(*)	(*)	4.68	(*)	(*)	8.00	8.74		5
1	viral load (log₁₀ GE/g) foetal thymus	(*)	(*)	6.54	7.40	7.54	7.00	6.34	7.41	6.34	(*)									7.67	(*)	7.89	6.79	6.26	(*)	7.08	5.85	7.05	1
2		(*)	(*)	(*)	(*)	5.94	(*)	(*)	8.31	6.74	7.78	(*)	(*)	(*)			(*)	4.88	(*)	9.33	8.40	7.72	5.27	(*)	(*)	(*)	(*)	(*)	2
3		(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	8.76						8.07	8.72	(*)	4.89	(*)	(*)	(*)	(*)	5.35	3
4		7.01	(*)	8.49	(*)	8.02	(*)	8.54	(*)	8.86										6.19	7.24	8.43	(*)	(*)	(*)	8.11	8.32	5.79	4
5		(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)										(*)	(*)	(*)	(*)	(*)	(*)	8.13	8.61	5
1	viral load (log₁₀ GE/mL) tongue fluids	(-)	(-)	5.26	4.94	6.27	5.03	6.68	(*)	4.22	5.09									5.72	(*)	6.31	4.96	4.73	(*)	5.05	(-)	4.62	1
2		(*)	(*)	(*)	3.36	(*)	(*)	(*)	7.05	(*)	5.43	(*)	(*)	(*)			(*)	3.75	(*)	5.75	4.87	4.94	(-)	(*)	(-)	3.37	(*)	(*)	2
3		(-)	3.62	(*)	(*)	(*)	(*)	(*)	(*)	(-)	(*)	(*)	(*)	6.69						4.55	5.36	3.58	3.67	(-)	(*)	(*)	(*)	(-)	3
4		(-)	3.88	5.46	(-)	(*)	4.53	4.94	(*)	(*)										(-)	(-)	4.28	(-)	(*)	4.00	(-)	(-)		4
5		(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)	(*)										(*)	(*)	(*)	5.39	(*)	(*)	4.23	4.41	5
1	viral load (log₁₀ GE/mL) tongue fluid-pool													4.96															1
2														4.06															2
3														4.95															3
4														8.19															4
5														3.98															5
1	viral load (log₁₀ GE/mL) processing fluid-pool													6.54															1
2														6.41															2
3														6.08															3
4														8.32															4
5														5.91															5

Table 1. *Foetal preservation status (VIA= viable; MEC= meconium stained; DEC= decomposed; AUT= autolysed) and viral load in serum, thymus and tongue fluids of individual foetuses of five infected gilts (1-5) as well as viral load in litter-wise pooled tongue fluids and processing fluids of the same litters. Viral load is displayed in genome equivalents per mL or g.*

Conclusion

① Under experimental conditions:

Positive correlation of viral loads in:

- Foetal serum and tongue fluids at the individual animal level.
- Foetal thymus and tongue fluids at the individual animal level.

② Field study:

Tongue fluids pooled by litter which were examined by RT-qPCR delivered positive results in the field study.

Compared to other sample material, tongue fluids seem to be a suitable sample material for PRRSv- monitoring in the field.

Positive correlation of viral loads in litter-pooled processing fluids and likewise litter-pooled tongue fluids.

Further reading

↗ *Porcine Reproductive and Respiratory Syndrome Surveillance in breeding Herds and Nurseries Using Tongue Tips from Dead Animals.*

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Lintjeshof veterinary practice,
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Eric van Esch.
Merefelt Livestock Diagnostics
the Netherlands

First field evaluation of an innovative tool for systematic PRRSv control – including a modified Holtkamp system – on farms under western European circumstances

Current problem

PRRSv is endemic in the Netherlands. The Western European swine industry has specific challenges for implementing a systematic approach, such as: the farm set-up, swine density and farm ownership structure.

Study objective

The objective of this study is to determine by a field evaluation if a proposed set of tools leads to a farm specific intervention plan directed towards PRRS control. The tool set is a combination of the following elements:

1. Monitoring and classification by a modified Holtkamp system.
2. Biosecurity check.
3. Decision making tool.

Study design

10 farms in the Netherlands (in total 20.000 sows) participated in the study. A combination of 3 tools was used: monitoring and classification by a modification of the original and updated Holtkamp system (1,2) to better fit the European circumstances, a biosecurity check (BioCheck UGent) and a decision making tool. The monitoring system is a diagnostic screening method to classify the farms' PRRS status as either unstable (red), stable with presence of PRRSv field strain in nursery pigs (orange) or stable without any PRRSv fieldstrain (green).

Results

The monitoring system showed that six out of ten farms classified as red at some point during the trial. None of the farms showed a consistent green status. The biosecurity checks showed that the weakest element of external biosecurity was the location of the farms because 80 % of the farms are located in swine dense areas. The weakest elements of internal biosecurity were: movement of piglets between litters, hygiene in piglet handling procedures, sick piglet handling, use of separate materials between age groups and cleaning and desinfecting of materials and boots. The most advised farm specific management interventions can be categorised as: prevention of contact between age groups, hygiene and piglet management in the farrowing unit and gilt introduction. Conclusion: On all farms the combination of the 3 tools lead to a farm specific plan. Subsequently a part of the advice in the plans was actually implemented, in varying degrees between participating farms. Restrictions for not implementing were mainly due to: challenges with staff, time and internal farm design.

Practical benefit

The field evaluation of a novel PRRS control tool can highly benefit the Western European swine industry, because this practical instrument can be the driver to start PRRSv control on farms.

Further reading

• Koenders et al., ESPHM 2023

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SEGES, Danmark

Gitte Drejer.
Danvet, Denmark

Managing the PRRS positive sow herd – breeding stock introduction and biosecurity

Study objective

The main objective with the present study is to establish whether it is possible to maintain a stable condition in an endemically infected sow herd with PRRS virus without mass vaccination of the sows with PRRS MLV vaccine. A stable condition is defined as a herd with no transmission of PRRS virus in the farrowing unit (no PRRS virus positive newborn piglets). The main hypothesis, H1, is that it is possible to maintain a stable sow herd with an endemic infection with PRRS without the use of any PRRS MLV vaccine in the sow herd. The secondary hypothesis, H2, is, based on experience from the field, that a stable sow herd can only be maintained by using off-site quarantine with 2 administrations of PRRS MLV vaccine for introduction of breeding stock and sectionized farrowing units.

Summary

Longitudinal sampling of processing fluid (PF) and oral fluid (OF) for PRRS PCR analysis was performed in herds that did or did not follow the general Danvet recommendations for an off-site quarantine and PRRS vaccination of new breeding stock. In the study, we used longitudinal monitoring with PCR analysis of Processing fluids (PF) from litters born from 1st litter sows or older sows (> 6th litter), because those were most likely to have reduced antibody levels, get infected with PRRS virus and transmit the virus to their offspring. Monitoring was done during 13 weeks in the winter season in herds not using mass vaccination with PRRS MLV vaccine.

When the sampled batches of pigs were weaned, OF was collected in the nursery 1-3 days after weaning, with piglets from different parities mixed in the same pens.

Information about biosecurity measures was collected with a questionnaire (22 questions).

All samples were analysed on the herd level for PRRS viral genome by PCR analysis, with pooling of PF from not more than 20 litters per analysis. OF was analyzed separately for each weekly batch. The samples were analyzed with a commercially available PCR test from an accredited, Danish laboratory in cooperation with Professor Lars E. Larsen, Danish Technical University, Copenhagen, DK.

Results

Four herds were included in the study, 2 herds using an off-site quarantine (group 1) and 2 herds without a regular quarantine facilities (group 2) (table 1).

PRRS virus was not detected in any of the samples collected for 13 weeks in the study herds, neither the herds with an off-site quarantine with PRRS-vaccination of new breeding stock or herds not using a regular quarantine. With the intensive sampling protocol used, we think that we beyond reasonable doubt showed that there was no circulation of PRRS virus in the farrowing units in these 4 herds.

2021

Managing the PRRS positive sow herd – breeding stock introduction and biosecurity

Table 1. *Production in 4 PRRS type 1 positive herds*

Herd No.	1	2	3	4
Study group	1	1	2	2
# sows	2860	1400	975	970
Avg. # liveborns/litter	19,5	18,1	17,2	18,5
Avg. # dead borns /litter	1,7	2,1	1,7	1,7
Avg. # weaned pigs /litter	16,5	15,9	14,6	16,4
% dead before weaning	14,5	13	14,2	8
Time since PRRS MLV				
Mass vaccination	2 years	5 years	3 years	8 months

Studu group 1:
Gilt introduction off-site quarantine with 2 x PRRS MLV vacc.
Study group 2:
Gilt introduction not separated from sow unit, no PRRS vacc.
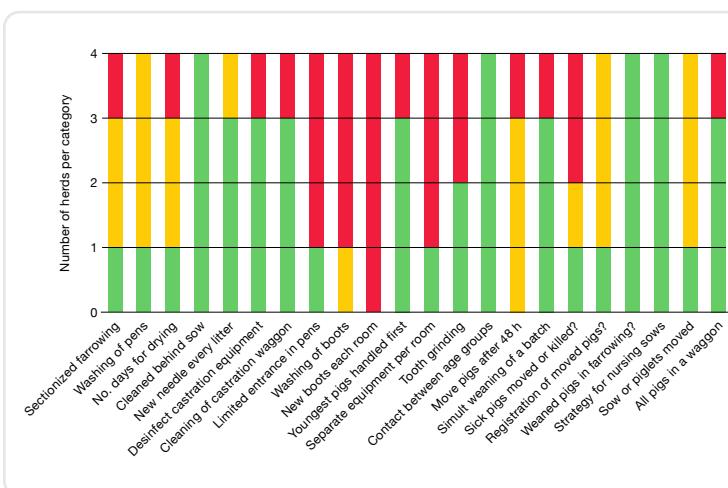
Figure 1. *Adherence to biosecurity recommendations.*
Green: *Complete adherence;* ***Yellow:*** *Partly adherence;*
Red: *Procedures not according to vet. biosecurity advice*

Hanne Bak.
SEGES, Denmark

Gitte Drejer.
Danvet, Denmark

Discussion and Conclusion

This study shows that it is possible to maintain a stable PRRS type 1 positive sow herd that weans virus negative pigs without the use of mass vaccination in the sow herd. Also, introduction of breeding stock via a quarantine facility with vaccination with PRRS MLV vaccine was not necessary to maintain a stable sow herd, because 2 of the 4 herds did not have a quarantine facility. Among the recorded management procedures, the following were applied in all 4 stable sow herds and can be recommended for maintenance of a stable sow herd: No contact between age groups, no weaned pigs in the farrowing unit, using a recommended strategy for nursing sows (2-step strategy) and daily cleaning behind the sow. The results obtained in this study might be biased by the fact that all study herds had been endemically infected with PRRS type 1 for several years. Therefore, herds infected more recently might still have to adhere to the Danvet protocol and possibly use a vaccination protocol to control the infection.

Practical benefit

The use of mass vaccination as a strategy to control PRRS in endemically infected sow herds has been studied repeatedly, but to our knowledge, no studies included a control group to test whether a non-vaccination strategy might be just as beneficial. In Denmark, most of the mass vaccinations with PRRS MLV vaccines in Denmark are done off-label due to the country-specific vaccine registrations, which entails problems with authorities and insurance companies. These would be avoided with an alternative protocol for herd stability.

2020

Award-winning projects:

PRRSv-1 recombination in swine herds: an emerging risk or a hype?

Erhard Van der Vries. GD Animal Health, the Netherlands

Impact of weaning procedures on PRRSv in the nursery section

Pia R. Heiselberg. HyoVet, Denmark

Biosecurity and management impact on PRRS Status and economical profit: Statistical process control after evaluation and improvement

Guillermo Ramis. Universidad de Murcia, Spain

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PRRSV-1 recombination in swine herds: an emerging risk or a hype?

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▶ **Episode: 16**   39 min.

2020 European PRRS Research Awards: Project leaders present their winning proposals

"It is feasible to do it in a year, but we are going to be busy!"

In 2020, Boehringer Ingelheim has sponsored its latest annual European PRRS Research Awards which offer 25,000 Euros towards the funding of each of three research projects that are potentially of practical benefit in controlling PRRS. Hear the winners of the 2020 Awards describe the research they propose, given the time limit of completing the work in one year. A practitioner-led project in Denmark is to examine the impact of piglet weaning strategies on PRRSv in the nursery. An international team led from The Netherlands will use whole-genome sequencing to investigate genetic recombination in type-1 PRRS viruses. And, a study in Spain aims to measure how biosecurity measures against PRRS relate to the physical performance and profitability of commercial swine farms.

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2020

PRRSv-1 recombination in swine herds: an emerging risk or a hype?

Background

Porcine reproductive and respiratory syndrome (PRRS) causes a significant economic burden to the swine industry. Recent evidence indicates that recombination events between different strains of PRRS virus type 1 (PRRSv-1) may play a role in European PRRSv-1 epidemiology, but existing genotyping methods fall short to study the incidence, spread and impact of these chimeric strains in the pig populations.

Aim

To develop a PRRS whole genome sequencing method using Oxford Nanopore Technology (ONT) and study PRRSv-1 recombination and epidemiology in the field.

Materials and methods

Complete PRRSv-1 ONT sequencing
A targeted PRRSv whole genome ONT sequencing methodology was developed. This was done in three successive steps. First, an existing open reading frame (ORF) 5 Sanger sequencing assay was transferred to the ONT-platform. Second, an ONT ORF 2-7 assay was developed by designing 3 sets of primer-pairs (n=31) to amplify the ORF 2-7 genome regions of all available PRRSv-1 EU sequences covered by tiled ~1,500 base pair fragments. (Figure 1). Third, additional primer-pairs were added to the ORF2-7 assay for amplification of 9 additional overlapping ~ 1,500 bp fragments covering the ORF 1 region and to allow complete PRRSv-1 genome sequencing (Figure 2).

Erhard Van der Vries.
GD Animal Health, the Netherlands

Figure 1. Scheme of PRRSv-1 ORF2-7 primer sets
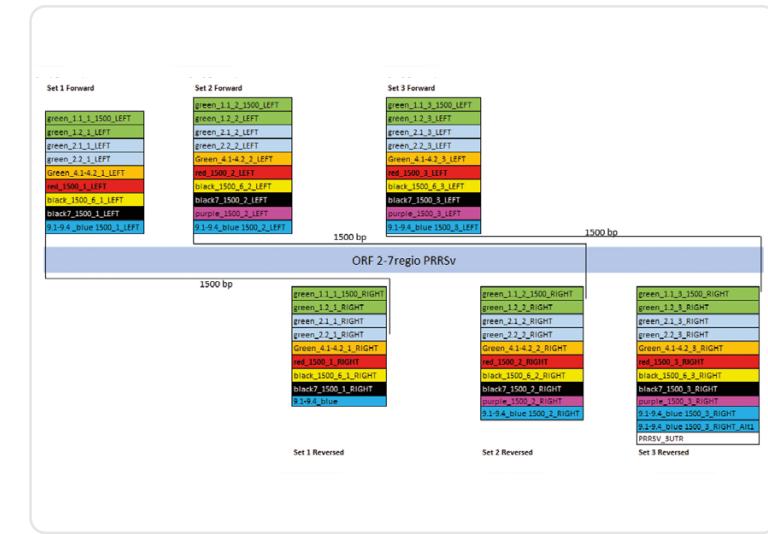
Sample collection
PRRSv-1 PCR-positive respiratory, serum and lung tissue samples (n=78) were collected from the GD repository. These were collected in the Netherlands and Eastern Europe between 2014 and 2016, some (n=12) of which contained a suspected recombinant PRRSv-1 virus strain. Between 2020-2021, additional oral fluid samples (n=88) were collected from pigs housed on farms (n=24) in the Netherlands. On some of these farms (n=12) a suspected recombinant strain was detected previously. PRRSv infection status and epidemiology data were recorded, including farm type, size, vaccination status and pig movements (data not shown here).

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# PRRSv-1 recombination in swine herds: an emerging risk or a hype?

Data analyses

A bioinformatics pipeline was built for automated 1) quality control of the reads, 2) generating a sequence consensus. Additionally, a customer report was generated, showing %-identity scores to existing vaccine strains for each ORF. Phylogenetic and recombination analyses was done using MrBayes and Dualbrothers recombination detection software available within the Geneious Prime software package (version 11.0.15). Practical benefit

Results

1 Both ORF-5 and ORF2-7 ONT sequencing methods were established successfully with a high success rate of achieving complete consensus sequences from clinical samples (n=79; median Ct=29) as compared to the traditional Sanger sequencing method. ORF 5 (ONT 90% versus Sanger 84%).

Figure 2. *The PRRSv genome. PRRS virus genome consists of at least 7 open-reading frames encoding for several structural and non-structural virus proteins.*
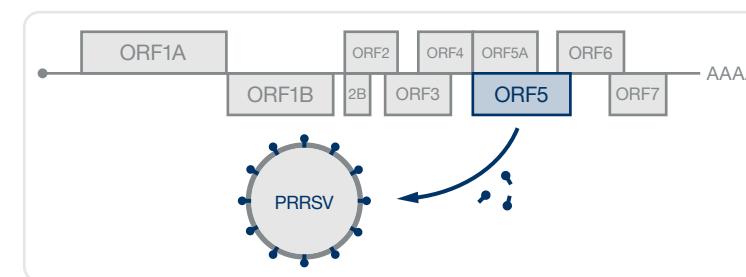
2 The development of the complete WGS method resulted in a much lower hit rate and average sequence coverage. This approach needs further optimization (Figure 3).

3 Unfortunately, no ORF2-7 or complete PRRSv-1 consensus sequences could be obtained from virus-positive animals on the farms with a suspected PRRSv-1 recombinant virus between 2014-2016. Several obstacles were: privacy-regulations to contact the farms (NL), outbreaks of African Swine Fever (POL) and SARS-COV-19, low virus loads (Ct values >30).

4 No signs of recombination could be found in the ORF 2-7 consensus sequences obtained from viruses in respiratory samples (77/88; 85%) collected within the 2020-2021 sampling period.

Figure 3. *Coverage plots for ORF2-7 (A) and (B) WGS PRRSv-1 sequencing methods*
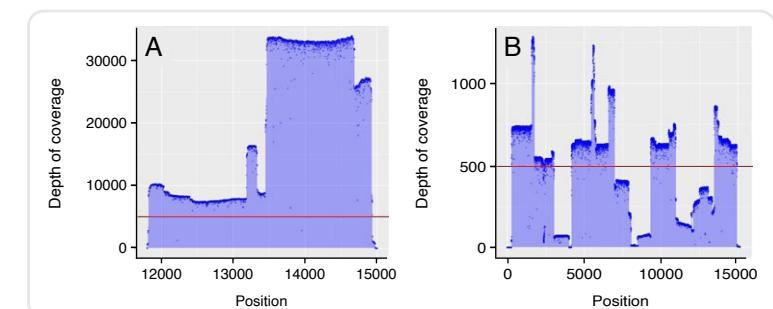
Discussion and conclusion

• ORF-5 and ORF2-7 ONT sequencing was successfully implemented. Additional work needs to be done to reliably obtain WGS consensus sequences from PRRS-1 positive samples.

• No evidence was found for any PRRSv-1 recombination events within the 2020-2021 period.

• In depth analyses of recombination events require better analyses tools and more available complete PRRSv-1 sequences.

Pia R. Heiselberg.
HyoVet, Denmark

Impact of weaning procedures on PRRSv in the nursery section

Current problem

During recent years there has been an increasing public pressure for reducing the use of antibiotic in Danish pig herds, and many producers therefore thrive to increase the health of pigs also in the nursery period. A vast number of publications describes protocols for obtaining stability in the sow herd, however, there is a lack of data that describe the variations between different weaning strategies on the downstream circulation of PRRSv in the period from weaning until 30 kg.

Study objective

The overall aim of the project was to compare the circulation and impact of PRRSv in piglets from weaning to 30 kg in Danish herds practicing two different weaning strategies.

Summary

The impact of different weaning strategies on the downstream circulation of PRRSv has not been widely described. In this study, piglets were sampled in three herds that performed "mixed at weaning (MIX)" and three herds that performed "all in/all out at weaning (AIAO)". Oral fluid samples were collected from four batches in each herd three times from weaning until 30kg and tested for PRRS virus and antibodies. Herds that used MIX at weaning had an eightfold increase in risk of detecting PRRSv in oral fluids. The level of PRRSv antibodies in oral fluid samples decreased in most of the batches that used AIAO whereas the opposite was the case for the MIX herds. In addition to oral fluid, tongue samples were collected from dead pigs and tested for PRRSv.

In 17 of the 23 batches the results of the tongue sample tests correlated with the results of the oral fluid sample tests.

Production data was also collected but could not clearly be related to the PRRSv status of the nursery sections albeit there was a tendency towards a lower mortality in the AIAO herds.

Overall, the results of the study confirmed that the weaning strategy has an impact on the circulation of PRRSv post weaning.

Practical benefit

Overall, the results of the study confirmed that the weaning strategy had a clear impact on the circulation of PRRSv post weaning. Based on the findings of this project AIAO strategies should be emphasized when weaning pigs in order to reduce the amount of virus circulating in the nursery.

Further reading

↗ *Effect of reducing crossfostering at birth on piglet mortality and performance during an acute outbreak of porcine reproductive and respiratory syndrome.*

2020

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Biosecurity and management impact on PRRS Status and economical profit: Statistical process control after evaluation and improvement

Current problem

Porcine Reproductive and Respiratory Syndrome (PRRS) remains one of the major health and economic problems of the pig industry. With ever larger farms and densely populated areas, biosecurity is a key issue in the prevention and control of the disease. This becomes especially important when dealing with a virus with a very high mutation rate, subject to the emergence of highly virulent strains at any time. And vaccination alone is not enough to prevent the effects of the disease.

Project objectives and practical benefits

The objectives of the project were to evaluate by statistical methods the evolution of 12 farms in SE Spain after implementing changes in biosecurity (facilities and management) and to validate their effectiveness in order to be implemented in the whole company (more than 90,000 sows) and to publish them for the benefit of the whole scientific community. To this end, the level of biosecurity and its evolution, the improvement of the main production parameters and the economic return on investment were studied.

Study Setup

Biosecurity was assessed using COMBAT (BI, Germany) and points for improvement were studied. Changes were made to the facilities, mainly focused on limiting human and vehicle traffic, carcass and slurry handling, cleaning and disinfection, AI/AO, litter management (bagged boxes, fang filing, hygiene between litters), animal flows, staff training, etc. The investment and consequent return on investment derived from the number of animals brought to slaughter after improvements was calculated using the BECAL software (BI, Germany). For the data analysis, statistical process analysis and discriminant function analysis were used in order to combine all analysed parameters.

Results

Differences were found in piglets born alive per farrowing (LBW) on all farms comparing all 3 years, while in piglets weaned per sow per year there were differences on all farms comparing 2019 and 2020. There was a worsening of parameters such as weaning weight or pre-weaning mortality, but which are significantly related to the increase in piglets born. In 2020, 37,816 piglets were produced more than in 2019, which means that sent to slaughter, there was a return of 10.10:1. After doing the FDA, on most farms there was a clear difference between 2019 with 2020 and 2021, but not so pronounced when comparing 2020 with 2021. This suggests that the effect of biosecurity improvements is sustained over time.

2020

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Universidad de Murcia, Spain

Biosecurity and management impact on PRRS Status and economical profit: Statistical process control after evaluation and improvement

Discussion and Conclusion

Improvements in biosecurity are one of the pillars of disease control and prevention, which has an impact on production data (Dee *et al.*, 2004; Postma *et al.*, 2015). Recent studies stipulate an average loss attributable to PRRS of € 255/sow (Renken *et al.*, 2021), so losses in a structure such as the one analysed would be of € 4,033,080 in 2020. However, the excess pigs sent for slaughter in 2020 compared to 2019 from the farms involved produced a profit of € 6,072,691, 384 per sow; much higher than described above.

In conclusion, the biosecurity improvements implemented on the farms studied have improved most of the production parameters and consequently the economic performance. Some parameters worsen as they are negatively correlated with the most economically important ones, so they cannot be analysed in isolation.

Figure 1. Variations in internal, external, management location and global risks, taking data from all farms and comparing before and after the intervention. These were calculated using COMBAT (BI). Significance is shown in red for independent sample comparisons and in blue for paired samples.
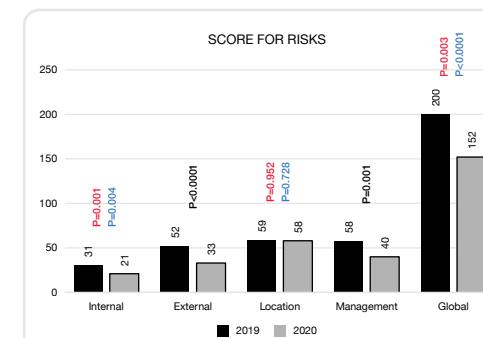
Figure 2. Example of SPC approach for farm D for the parameters piglets born alive per farrowing (ABP) and piglets weaned per sow per year (PWSY). Below the dot plot obtained in the calculation of discriminant functions comparing all the parameters analysed in the study period.
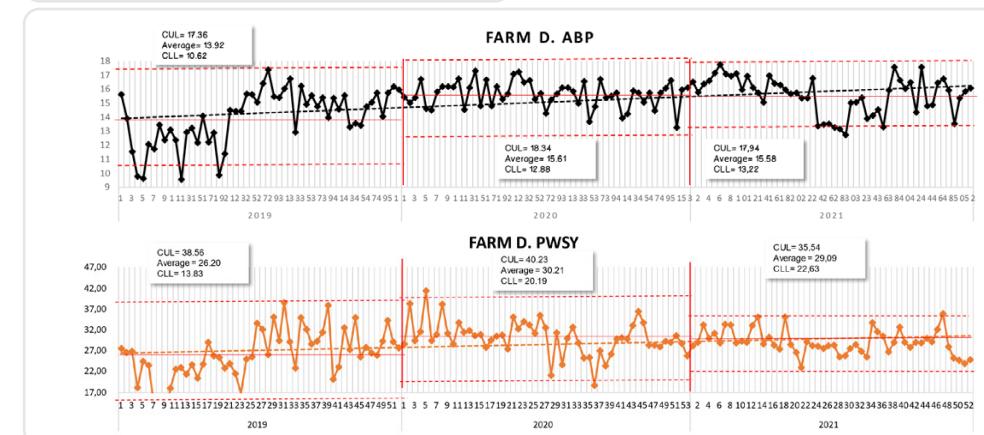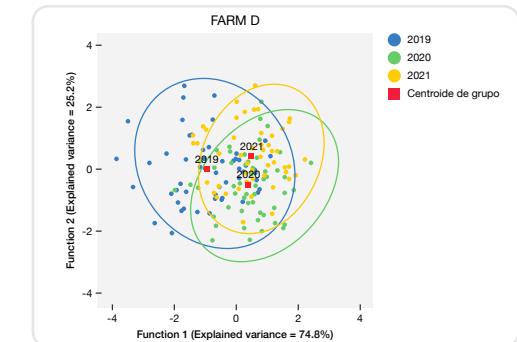
2019

Award-winning projects:

Role of cytotoxic T lymphocytes in gilts after ML vaccination in protection against vertical transmission of PRRSv

Yanli Li. Universitat Autònoma de Barcelona, Spain

Impact of repeated vaccinations with MLV PRRSv vaccines on the adaptive immune response

Lars Erik Larsen. University of Copenhagen, Denmark

Molecular traceability of PRRSv: an epidemiological tool for improving biosecurity

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European**PRRS**
R E S E A R C H
AWARD

2019

Yanli Li.
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Role of cytotoxic T lymphocytes in gilts after ML vaccination in protection against vertical transmission of PRRSv

Current problem

Given the diversity of PRRSv strains, vaccination is considered to produce only partial protection. The protection is thought to be mediated in part by neutralizing antibodies (NA), generally with low titers and only specific for the strain that induces them. Cell-mediated immunity (CMI) is considered as the second factor since high frequency of PRRSv-specific IFN-γ secreting cells (IFN-γ-SC) has been proved to be related with protection in piglets. The development of cytotoxic T lymphocytes (CTL) is thought to play a crucial role in eliminating infected cells in most of viral infections. However, up to now, data about the role of CTL during PRRSv infection and the induction as a result of vaccination are very limited.

Study objective

To determine the development of cytotoxic T lymphocytes (CTL) in gilts after MLV vaccination and the relationship of CTL and neutralizing antibodies levels with the probability of vertical transmission of PRRSv.

Summary

Porcine reproductive and respiratory syndrome virus (PRRSv) is one of the major swine pathogens causing reproductive failure in sows. Although modified-live virus (MLV) vaccines are available, only partial protection against heterologous strains is produced, thus vaccinated sows can be infected and cause transplacental infection. The immune effector mechanisms involved are largely unknown.

The present study investigated the role of cytotoxic lymphocytes, including cytotoxic T cells (CTL), NKT, and NK cells, in preventing PRRSv1 transplacental infection in vaccinated primiparous sows (two doses vaccinated). Sows from a PRRSv1 unstable farm were bled just before the last month of gestation (critical period for transplacental infection), then followed to determine whether sows delivered PRRSv1-infected (n=8) or healthy (n=10) piglets. After that, we compared functions of CTL, NKT, and NK cells in two groups of sows. No difference was found through cell surface staining. But upon in vitro re-stimulation with the field circulating virus, sows that delivered PRRSv1-free piglets displayed a higher frequency of virus-specific CD107a+ IFN-γ-producing T cells, which accumulated in the CD4+ compartment including CD4 single-positive (CD4 SP) and CD4/CD8γ double-positive (CD4/CD8γ DP) subsets. The same group of sows also harbored a higher proportion of CD107a+ TNF-γ-producing T cells that predominantly accumulated in CD4/CD8γ double-negative (CD4/CD8γ DN) subset. Consistently, CD4 SP and CD4/CD8γ DN T cells from sows delivering PRRSv1-free piglets had higher virus-specific proliferative responses. These data strongly suggest that CTL responses correlate with protection against PRRSv1 transplacental infection, being executed by CD4 T cells or CD4/CD8γ DN T cells.

Role of cytotoxic T lymphocytes in gilts after ML vaccination in protection against vertical transmission of PRRSv

Practical benefit

The present research correlates the CTL and NA in gilts with vertical transmission, which is more related to the field swine industry, especially when a widely used MLV is combined. Besides vertical transmission, the present proposal will also examine the development of CTL among newly introduced gilts. This can provide a guidance for the future vaccination protocols and vaccines, for instance, investigation of adjuvants to promote higher CTL responses, or modification of vaccination schedules to increase levels of CTL in the susceptible period of vertical transmission, etc.

Further reading

↗ *Cytotoxic T cells Correlate with Protection Against PRRSv1 Transplacental Infection. Frontiers in Immunology.*

Yanli Li.
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Lars Erik Larsen.
University of Copenhagen, Denmark

Impact of repeated vaccinations with MLV PRRSv vaccines on the adaptive immune response

Study objective

To investigate the impact of repeated vaccinations on the adaptive immune responses to homologous and heterologous strains of PRRSv.

Summary

The results of this study revealed that to achieve the full advantage of PRRSv sow mass vaccination, i.e., to obtain PRRS virus-free nursery units, compliance towards the basic rules for effective PRRSv control is required. No negative side effects of the MLV PRRS vaccination were seen, probably because the most vulnerable age groups were exempted from vaccination. The results provided valuable information to herd veterinarian and producers on the benefit and limitation on mass-vaccination against PRRSv.

Practical benefit

Mass vaccination against PRRSv are widely used in pig herds also in Europe as a central part of the PRRSv control. The study will contribute to our understanding on the value of this strategy and provide information on the beneficial and potential detrimental impact of the procedure and by that support future adjustment of the vaccination strategies to the benefit of the producers.

Further reading

↗ *Impacts of Quarterly Sow Mass Vaccination with a Porcine Reproductive and Respiratory Syndrome Virus Type 1 (PRRSv-1) Modified Live Vaccine in Two Herds.*

2019

Gerard Eduard Martin Valls.
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Spain

Molecular traceability of PRRSv: an epidemiological tool for improving biosecurity

Current problem

PRRSv is considered one of the costliest diseases for the pig industry. A successful control of PRRS requires a multiple approach including at least a biosecurity plan, a diagnostic and monitoring scheme, an immunisation protocol and the management of the pig flow. Biosecurity is key to reduce the risk of virus introduction in the farm. Usually, the prioritisation of biosecurity measures is mostly based on the general knowledge about how the disease is transmitted, on common sense and on the previous experiences of the veterinarian and the farmer. A precise evaluation of the potential effectiveness of different biosecurity measures would be an important help to decide what to do.

Study objective

The aim of the present project is to characterize the flow of PRRSv in a pyramidal system, using whole genome sequences obtained by NGS, as a tool to determine the transmission routes among farms. This data may contribute to assess the importance of the different routes and, consequently to establish a scientific method to prioritize biosecurity measures.

Summary

As far as recombination may occur, ORF5 sequencing may have a limited use for assessment of lateral introductions and genetic evolution of PRRSv. The objective of this study was to use whole genome sequencing for assessing PRRSv transmission between farms from a same company.

The results demonstrated that apparently similar strains may be recombinant. Also, in the evaluated farms a highly pathogenic PRRSv1 strain was detected. Whole genome characterization helped to evaluate differences with other wild type PRRSv1 strains circulating in the same company.

Practical benefit

This study demonstrates that whole genome sequencing is a powerful tool for evaluating PRRSv1 epidemiology. Also shows that ORF5 sequencing, although it is useful, has some limitations that can be overcome by whole genome sequencing. Finally, the present work helped to evaluate the efforts on biosecurity of the company that participated in the study, and to compare the impact of genetically different PRRSv1 strains during long periods of time.

Further reading

↗ *Introduction of a PRRSv-1 strain of increased virulence in a pig production structure in Spain: virus evolution and impact on production.*

2018

Award-winning projects:

➡ **Active surveillance of porcine productive and respiratory syndrome virus in breeding herds, nurseries and finishers from carcasses**

Jordi Baliellas. Grup de Sanejament Porcí, Spain

➡ **Interference of swine influenza virus infection with PRRS MLV vaccination in piglets**

Olivier Bourry. Anses, France

➡ **Is ORF5 nucleotide sequence analysis sufficient for tracing PRRSv-1 strains?**

Jos Dortmans. GD Animal Health, the Netherlands

Jordi Baliellas.
Grup de Sanejament Porcí, Spain

Active surveillance of porcine productive and respiratory syndrome virus in breeding herds, nurseries and finishers from carcasses

Current problem

One of the main issues regarding surveillance and monitoring of large pig populations is a) how to detect PRRS in low prevalence scenarios and b) how to diagnose and monitor it in a cost effective and convenient way. New sampling protocols and specimens are needed, that address these two topics.

Study objective

As castration is no longer allowed in Europe for animal welfare reasons can a risk based sample such as tongue tip samples from dead animals replace currently used specimen and detect PRRS in low prevalence scenarios?

Summary

Over the last years, new techniques based on the aggregated sampling concept have emerged as an alternative to the use of serum to monitor PRRSv. For example with the use of oral fluids and processing fluids we monitor: in a cost-effective way, more animals more frequently and it is usually non-invasive and convenient. The use of the fluids from tongue tips of dead animals in a farm (TTF) is a new possibility to monitor PRRSv at birth (tongue tips of stillborn piglets), during the suckling phase, nursery or finishing period (tongue tips of dead animals in each stage). We collect these tongue tips in aggregated bags, we store them in frozen conditions and during the thawing process we obtain a liquid used to carry out a PCR or to sequence the positive PCR samples. After the first studies we conclude that TTF has a good PRRSv detection capacity and it is a feasible technique to put into practice for continuous monitoring.

Study results

- The agreement between tongue exudate and serum was good. The main discrepancy came from positive samples in tongue exudate and not in serum (carriers and low prevalence scenarios).

- Low cT values of PCR positives samples facilitates sequencing.

- ORF5 sequences obtained from TTF were genetically similar to sequences obtained from serum.

Practical benefit

- TTF is an easy, cheap and welfare friendly PRRSv monitoring technique.

- TTF is an alternative to processing fluid in farms where castration is applied.

- Low cT values in TTF allows to obtain the PRRSv sequence and to predict if the farm is close to the stability with continuous monitoring.

Further reading

↗ *Porcine Reproductive and Respiratory Syndrome Surveillance in breeding Herds and Nurseries Using Tongue Tips from Dead Animals.*

Olivier Bourry.
Anses, France

Interference of swine influenza virus infection with PRRS MLV vaccination in piglets

Current problem

Modified-live vaccines (MLVs) against PRRSvs are usually administrated to piglets at weaning when swine influenza A virus (swIAV) infections frequently occur. SwIAV infection induces a strong interferon alpha (IFNa) response and IFNa was shown to abrogate PRRSv2 MLV replication and an inherent immune response. To the authors knowledge, no information is available on potential negative effects of an ongoing swIAV infection on PRRS MLV vaccine efficacy.

Study objective

Evaluate the impacts of swIAV infection on the replication of a PRRSv MLV, post-vaccine immune responses and post-challenge vaccine efficacy at both the systemic and pulmonary levels.

Summary

Piglets were either swIAV inoculated and MLV1 vaccinated 6 h apart or singly vaccinated or mock inoculated and mock vaccinated. Four weeks after vaccination, the piglets were challenged with a PRRSv1 field strain. The results showed that swIAV infection delayed MLV1 viremia by six days and post-vaccine seroconversion by four days. After the PRRSv1 challenge, the swIAV enhanced the PRRSv1-specific cell-mediated immunity (CMI) but the PRRSv1 field strain viremia was not better controlled. High IFNa levels that were detected early after swIAV infection could have been responsible for both the inhibition of MLV1 replica-

tion and CMI enhancement. Thus, whereas swIAV infection had a negative impact on humoral responses post-vaccination, it did not interfere with the protective effectiveness of the PRRSv MLV1 in our experimental conditions.

Practical benefit

Understanding a potential impact of Swine Influenza infections on PRRS MLV vaccination, will help to maximize the benefit of vaccination and avoid potential interferences.

Further reading

↗ *Concomitant Swine Influenza A Virus Infection Alters PRRSv1 MLV Viremia in Piglets but Does Not Interfere with Vaccine Protection in Experimental Conditions.*

2018

Jos Dortmans.
GD Animal Health, the Netherlands

Is ORF5 nucleotide sequence analysis sufficient for tracing PRRSv-1 strains?

Current problem

In regional elimination and national eradication efforts PRRSv genotyping is one of the key tools to assess the performance of the action and to help improving internal and external biosecurity measures, and to better understand the virus ecology. PRRSv genotyping is being performed mostly based on ORF5 and/or ORF7 sequence analysis. Based on the sequence of ORF5 alone the identity of the strain of interest would be misinterpreted and wrong conclusions may be drawn in a diagnostic and epidemiological perspective. When the source of a particular strain is incorrectly determined as either a new introduction or recurring internal circulation, this may result in an incorrect advice regarding biosecurity measures.
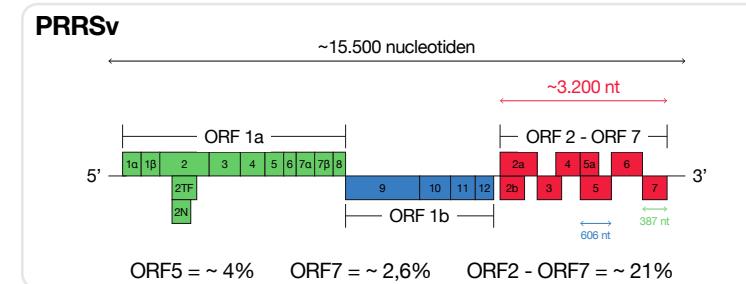
Picture PRRS genome: ORF 5 only reflects a minor part of the entire PRRS genome (~4%)

Study objective

Unfortunately, unlike in PRRSv-2, the knowledge about PRRSv-1 recombination frequency and recombination hot spots is largely missing. The objective of this study is to investigate PRRSv-1 recombinations based on ORF2-7 sequences.

Summary

Thirty-eight PRRSv-1 sequences of ORF2-ORF7 from the Netherlands, as well as 84 PRRSv-1 sequences from Europe, Asia and America, available in GenBank were aligned and analyzed using the RDP4 program to detect potential recombinant viruses in the dataset.

Analysis showed 57 sequences with some recombination evidence. Recombination in 30 sequences was detected by most algorithms incorporated in RDP4 programs. The majority of the detected recombination events were unique and at random positions. In some cases the analysis showed that the position in the phylogenetic tree topologies was OF dependent, supporting genetic recombination in their emergence. Interestingly, Dutch sequence NL/GD-5-18/2015 clustered with the highly virulent Austrian strain AUT/15-33/2015 in phylogenetic trees constructed from complete ORF2, ORF3, ORF4 and ORF5 nucleotide sequences, whereas in the ORF6 and ORF7 trees it clustered with Lelystad virus.

Our results provide new insights into the role of genetic recombination in PRRSv-1 evolution. Furthermore, it will allow to better assess the value and limitations of ORF5 sequence analysis in epidemiological investigations.

Further reading

• *Dortmans, ESPHM 2019, oral presentation*

2017

Award-winning projects:

➡ **OPTIVAC: Optimization of Porcine Reproductive and Respiratory Syndrome Virus (PRRSv) vaccination to enhance control of the virus in the field**

Valérie Normand. Porc.Spective, France

➡ **Identification of epitopes responsible for the induction of broadly neutralizing antibodies**

Cinta Prieto. Universidad Complutense de Madrid, Spain

➡ **Assessment of the vertical transmission of PRRSv1 in unstable farm: effect of parity and neutralizing antibody titers**

Jordi Soto Vigueras. Universitat Autònoma de Barcelona, Spain

OPTIVAC: Optimization of Porcine Reproductive and Respiratory Syndrome Virus (PRRSv) vaccination to enhance control of the virus in the field

Valérie Normand.
Porc.Spective, France

Current problem

As one of the most costly swine disease PRRS control is equally as important and challenging. One key aspect of PRRS control is vaccination. Optimizing these vaccination protocols can help to control PRRS.

Study objective

The protection elicited at the end of the post-weaning period may be the key to stop PRRSv circulation and therefore to achieve an inactive status of the fattening unit. In the field, vaccination protocol should be scheduled in order to take into account maternal derived immunity (MDA) impact and to enable piglet's protection with low or high level of MDAs at the time of vaccination. Thus, the aim of the current field study is to investigate the influence of different piglets PRRS MLV vaccination protocols on the development of post-vaccination humoral and cellular immune responses, and then on the vaccine efficacy regarding viral circulation and clinical improvement.

Summary

This longitudinal study was the first evaluation, under field conditions, designed to assess different PRRSv MLV vaccination protocols on (PRRSv-NAs) interference: Group A (Control), Group B (Vaccinated at 7 weeks of age), Group C (Vaccinated at 3 weeks of age), Group D (Vaccinated at 3 and 7 weeks of age).

The aim was to confirm kinetics of PRRSv-NAs decay, and the vaccine efficacy regarding immune response, viral circulation and clinical improvement.

Post-vaccination virological & immune responses:

- We observed a higher heterogeneity of the immune response at 11 weeks in piglets vaccinated at 3 weeks old.
- We defined a cluster of pigs with the highest level of immune parameters on the vaccinated batch at 3 & 7 weeks old.

PRRSv field strain infection:

- We showed that the infection was earlier and more drastic in unvaccinated pigs vs. vaccinated pigs.
- There was no difference among vaccination program/ PRRSv infection dynamics in vaccinated pigs.

Cinta Prieto.
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Spain

Identification of epitopes responsible for the induction of broadly neutralizing antibodies

Current problem

To control PRRS, one of the most costly diseases in swine production, vaccines are commonly used. However, protection achieved by vaccination, and even by previous exposure to the virus, is often partial. Although other factors might be involved, the remarkable PRRSv variability, which leads to a high antigenic diversity and limited cross-reactivity between strains, is very likely responsible for the partial protection against secondary infections provided by primary infections and vaccination. Although the components of the immune response responsible for protection have not been definitively identified, it has been demonstrated that neutralizing antibodies (NA) might play a role in protection, at least against reinfections. Broadly reactive NAs have been described after infection by other viruses as influenza or human immunodeficiency virus (HIV), against which elite neutralizers develop broadly neutralizing antibodies and are protected against disease. This observation has allowed developing the theory that the response against those theoretically poorly immunogenic epitopes might be the key factors for protection.

For PRRSv, it is plausible that conserved neutralization epitopes may exist. However, no attempts have been made to identify conserved epitopes responsible for cross-neutralization, which would be the candidate antigens for the induction of broadly NA in vaccinated pigs.

Study objective

Identify and characterize conserved linear neutralizing epitopes in the PRRSv which are involved in development of broadly reactive NA by means of Pepscan analysis using sera of known reactivity and viruses which have been previously confronted to those sera.

To achieve the study objective, a total of 28 sera selected on the basis of their cross-reactivity and their origin and four viruses, selected on the basis of their reactivity with the selected sera will be used. Genes coding for GP2, GP3, GP4, GP5 and M of each of the selected viruses will be sequenced and the sequences used to design peptides which will be used in a Pepscan assay. The identification of conserved neutralizing epitopes is very relevant for the development of new vaccine products that potentiate the response against those particular epitopes. Besides, the identification of conserved neutralizing epitopes could allow developing new diagnostic assays, particularly ELISA assays which might be useful to predict the level of protection on the pig population or individual pigs.

↗ *The Ability of Porcine Reproductive and Respiratory Syndrome Virus Isolates to Induce Broadly Reactive Neutralizing Antibodies Correlates With In Vivo Protection.*

2017

Jordi Soto Vigueras.
Universitat Autònoma de Barcelona,
Spain

Assessment of the vertical transmission of PRRSv1 in unstable farm: effect of parity and neutralizing antibody titers

Current problem

PRRS was first described in the United States in 1987 and since then has become the most costly among the common diseases of pig.

After the introduction of PRRSv1 in a farm the infection spreads in sows that give birth to viremic piglets. These, will bring the infection downstream to the nurseries and growing units. Most farms will become endemic as far as the virus continues circulating in the breeding herd they will remain so. During this endemic state vertical transmission occurs causing the perpetuation of the infection in nurseries. These farms where the virus circulates in breeders and produce viremic piglets at weaning are designated as unstable.

It is unclear if circulation of the virus within the sows' stock is more frequent in younger or older sows or if the risk of circulation can be predicted based on the levels of antibodies. This is particularly important in vaccinated farms where, in principle, vaccination protocols are designed to provide efficient protection to gilts and to maintain immunity in sows.

Study objective

Determine if the age of the sow (young sows vs. old sows) and the level of neutralizing antibodies may be correlated with the occurrence of vertical transmission of PRRSv1 in vaccinated farms.

2016

Award-winning projects:

Field study to assess vertical transmission of type 1 strains of PRRS virus using pre-weaning oral fluid samples

Arnaud Lebret. Porc.Spective, France

Genetic programming of porcine memory B cells to enable the isolation of PRRSv-neutralizing monoclonal antibodies

Simon Graham. The Pirbright Institute, UK

Porcine Reproductive and Respiratory Syndrome virus (PRRSv): A Cross-Sectional Study on ELISA Seronegative, Multivaccinated Sows

Ann Brigitte Cay. Department of Infectious diseases in animals, Belgium

EuropeanPRRS
RESEARCH
AWARD

2016

Arnaud Lebret.
Porc.Spective, France

Field study to assess vertical transmission of type 1 strains of PRRS virus using pre-weaning oral fluid samples

Current problem

Defining shedding and exposure status for PRRSv is essential in herd stabilisation protocols and weaning-age pigs is a key subpopulation. Oral fluid (OF) sampling is a welfare-friendly and cost saving promising alternative but data comparing against serum samples are lacking.

Study objective

The first objective of our study was to compare the rate of detection of PRRSv-1 in individual serum sample, individual OF sample, litter-based OF sample, collected the day before weaning. The second objective was to evaluate the interest of pooling samples.

Summary

The study was performed on a 210-sows, PRRSv-1 exposed, with confirmed shedding, non-vaccinated against PRRSv, herd. 80 litters were sampled and 26 were viropositive and therefore included. The rate of detection of PRRSv-1 with RT-qrtPCR in blood samples, iOF and cOF was 67, 23 and 77%, respectively. The Ct values from RT-qrtPCR on collective OF were statistically lower if the serum of the piglet of the litter was positive. The lower the Cycle threshold (Ct) value of RT-qrtPCR on collective OF, the higher the probability that the serum sampled in the same litter was positive. Ability to detect PRRSv RNA after pooling was 67% for sera and 58% for cOF.

Conclusions

The rate of detection of PRRSv-1 was about the same in cOF and blood samples. Virus sequencing, if required, should be performed on individual serum samples. The smaller the Ct of a cOF sample from a litter, the greater the likelihood that the serum sample from a piglet of that litter is positive. A cost-effective and representative sampling protocol to monitor sow herds stabilisation of a sow batch could be: to collect both cOF and one serum sample per litter; to perform firstly RT-qrtPCR on pooled cOF; in case of negative results to consider the batch negative; in case of positive results in a unvaccinated herd or a killed vaccine vaccinated one to consider the batch positive; in case of positive result in a herd vaccinated with a modified live vaccine serum samples of litters with positive cOF should be tested for sequencing (selecting the litters with the lowest Ct for cOF).

Practical benefit

Current monitoring of herd status using exclusively serum sampling, takes time, can be stressful for both animals and personnel and painful for animals (especially young piglets). This study aims to help practitioners to optimize their sample collection (number/type/pooling) to investigate breeding herd status.

Further reading

↗ *Monitoring PRRSv-1 in suckling piglets in an endemic herd using reverse transcriptase quantitative real time polymerase chain reaction.*

2016

Simon Graham.
The Pirbright Institute, UK

Genetic programming of porcine memory B cells to enable the isolation of PRRSv-neutralizing monoclonal antibodies

Current problem

Current PRRSv vaccines provide limited protection; only providing complete protection against closely related strains. The development of improved PRRSv vaccines would benefit from an increased understanding of epitopes relevant to protection, including those recognized by antibodies which possess the ability to neutralize distantly related strains.

Study objective

In this work, a reverse vaccinology approach was taken; starting first with pigs known to have a broadly neutralizing antibody response and then investigating the responsible B cells/antibodies through the isolation of PRRSv neutralizing monoclonal antibodies (mAbs). PBMCs were harvested from pigs sequentially exposed to a modified-live PRRSv-2 vaccine as well as divergent PRRSv-2 field isolates. Memory B cells were immortalized and a total of 5 PRRSv-specific B-cell populations were isolated.

Conclusions

All identified PRRSv-specific antibodies were found to be broadly binding to all PRRSv-2 isolates tested, but not PRRSv-1 isolates. Antibodies against GP5 protein, commonly thought to possess a dominant PRRSv neutralizing epitope, were found to be highly abundant, as four out of five B cells populations were GP5 specific. One of the GP5-specific mAbs was shown to be neutral-

izing but this was only observed against homologous and not heterologous PRRSv strains. Further investigation of these antibodies, and others, may lead to the elucidation of conserved neutralizing epitopes that can be exploited for improved vaccine design and lays the groundwork for the study of broadly neutralizing antibodies against other porcine pathogens.

Practical benefit

This PRRS Award enabled the establishment of a system that can be used to isolate PRRSv-specific monoclonal antibodies from immune pigs. In addition to improving our understanding of the antibody response to PRRSv, these monoclonal antibodies will allow epitopes to be resolved that may ultimately guide the design of PRRS vaccines to induce cross-protective immunity.

Further reading

↗ *Isolation of Porcine Reproductive and Respiratory Syndrome Virus GP5-Specific, Neutralizing Monoclonal Antibodies From Hyperimmune Sows.*

2016

Ann Brigitte Cay.
Department of Infectious diseases
in animals, Belgium

Porcine Reproductive and Respiratory Syndrome virus (PRRSv): A Cross-Sectional Study on ELISA Seronegative, Multivaccinated Sows

Current problem

Vaccination against Porcine Reproductive and Respiratory Syndrome virus (PRRSv) is widely used to control clinical disease, but the effectiveness appears in some cases to be suboptimal. Field reports have stated the presence of routinely PRRSv-vaccinated but ELISA seronegative sows: the ELISA non-responders. The real extent of this phenomenon (prevalence—origin—consequences) was not yet investigated.

Study objective

In this study, the prevalence of ELISA non-responders was assessed by measuring PRRSv-specific antibodies in 1400 sows, originating from 70 PRRSv-vaccinating sow herds, using IDEXX ELISA (ELISA 1) and CIVTEST E/S ELISA (ELISA 2). Neutralizing antibodies (NAbs) were quantified in a virus neutralization assay.

Conclusions

Univariable logistic regression was used to identify herd risk factors for the presence of ELISA non-responders. The global prevalence of non-responders varied from 3.5% (ELISA 1) to 4.1% (ELISA 2), the herd-level prevalence was 40% and the within-herd prevalence ranged from 5% to 20% (ELISA 1) and from 5% to 30% (ELISA 2). The ELISA non-responders had significantly lower NAbs than the ELISA responders. Herds using the combination of one modified live vaccine and one killed vaccine had a signifi-

cantly reduced risk of having ELISA non-responders. A first assessment of the prevalence and possible consequences of ELISA non-responders has been provided by this study. The clinical importance, origin and underlying immunological mechanisms warrant further research.

Further reading

↗ *Porcine Reproductive and Respiratory Syndrome virus (PRRSv): A Cross-Sectional Study on ELISA Seronegative, Multivaccinated Sows.*

2015

Award-winning projects:

- **Alternative Sampling Methods in newborn Piglets for PRRS Diagnosis**

  Gerard Eduard Martin Valls. Universitat Autònoma de Barcelona, Spain

- **PRRSsos Project**

  Carlos Pineiro Noguera. PigCHAMP, Spain

- **Interference of Maternally Derived Antibodies with PRRS vaccine in piglets: impact on viral parameters and transmission**

  Olivier Bourry. Anses, France

Alternative Sampling Methods in newborn Piglets for PRRS Diagnosis

Study objective

The objective of this study was to test the suitability of umbilical cord (UC) sampling and ear vein swabbing (EVS) as alternatives to jugular vein bleeding (JVB) for the assessment of vertical transmission of porcine reproductive and respiratory syndrome virus (PRRSv). The conclusions were that UC testing was a faster and more sensitive alternative to JVB or EV for the detection of PRRSv in newborn piglets.

Summary

- There was a high detection rate for PRRS virus in newborn piglets.

- Obtaining umbilical cord specimens was faster than bleeding newborn piglets.

- Umbilical cords were useful for determining vertical transmission of PRRS.

- Umbilical cord sampling upholds pig welfare and is a suitable method for sampling.

Practical benefit

This study demonstrates that collecting UC is a fast and sensitive method for sampling newborns. It can be done by farmers with short training and could be very useful for monitoring vertical transmission.

Further reading

↗ *Testing of umbilical cords by real time PCR is suitable for assessing vertical transmission of porcine reproductive and respiratory syndrome virus under field conditions.*

Gerard Eduard Martin Valls.
Universitat Autònoma de Barcelona, Spain

Carlos Pineiro Noguera.
PigCHAMP, Spain

PRRSsos Project

Study objective

The project has three main objectives; all of them are focused on the understanding of the effect on PRRS control by means of proper farm staff education and training, measuring at the same time the impact of the implemented measures.

1 To assess the impact in main key performance indicators of an adequate farm staff education and training program on PRRSv control in commercial farms.

2 To monitor PRRS evolution and its relationship with the implementation of measures control in particular with farm staff movements to evaluate the level of control of PRRS which could be reached using awareness and training as principal tool.

3 To promote and reinforce the role and engagement of farm staff in biosecurity plans related to PRRS control the research and the application methods based on the potential repercussions of PRRS control by the farm staff.

Conclusions

- The project proposed and later defined a new way of working in biosecurity control and farm operations generating data about farm staff, visitors and vehicles movements, within and among farms, allowing generating alerts when biosecurity and operations rules are not respected, real-time monitoring of people and vehicles and deeper analytics including machine learning algorithms to prevent problems. The system allows an evolution to data driven culture of working and customized training based on errors and risks detected. Also,

the system demonstrated a better production when biosecurity and operations rules are respected (Black *et al,* 2020).

- The system, named Biorisk, allows moving from guessing to knowing and monitoring biosecurity and monitoring real time, with prompt interactions and customized education and training.

Practical benefit

This project is proposing a new approach to PRRS control complimentary to those already existing in the sector (vaccination and management practices mainly) since it is focusing on the role of farm staff and its proper training, as a critical factor in PRRS control. Another added novelty is the smooth integration of the Information and Communication Technologies (part of peoples' lives in other aspects) as a simple and cheap tool for PRRS control.

Therefore, it is expected a better understanding of this important factor as well as a higher engagement of workers in the control of a disease that in many occasions is out of their understanding delivering undesired risks and gaps many times undetected.

This approach, being always important, can be even more in familiar or farrow to finish farms that don't have continuous or good access to structured education and training programs as is more common in large producers.

Further reading

↗ *Association between different types of within-farm worker movements and number of pigs weaned per sow in U.S. Swine farms.*

Olivier Bourry.
Anses, France

Interference of Maternally Derived Antibodies with PRRS vaccine in piglets: impact on viral parameters and transmission

Current problem

Modified live virus (MLV) vaccines are commonly used to reduce the impact of porcine reproductive and respiratory syndrome (PRRS) but limited information on potential maternal interference is available.

Study objective

The first objective of the present project is to investigate the interference of maternally derived antibodies (MDA) with a PRRS MLV vaccine in piglets in terms of virological parameters and viral transmission. The secondary objective is to assess the effect of MDA on vaccine efficacy in terms of clinical and growth parameters.

Summary

Here, we evaluated the impact of maternally-derived neutralizing antibodies (MDNAs) on vaccine efficacy after PRRS virus (PRRSv) challenge. Piglets with low (A-) or high (A+) MDNA levels derived from a commercial pig herd were moved to experimental facilities to be vaccinated (V+) or not (V-) with a PRRSv-1 MLV vaccine at 3 weeks of age (woa). Because of unexpectedly low vaccine detection in A-V+ piglets post-vaccination (pv), all V+ piglets received a second vaccination at 4 woa. Five weeks (W5) pv, piglets were inoculated with a PRRSv-1 field strain to evaluate vaccine protection, and were mingled 24 h later with noninoculated piglets of similar immune status to assess viral transmission. Vaccine strain was detected at W2 pv in 69% and 6% of A-V+ and A+V+ piglets, and at W5 pv in 50% and 25% of A-V+ and A+V+ piglets, respectively. At W5

pv, 94% of A-V+ and 44% of A+V+ piglets seroconverted, with a significant IFNg response induction in the A-V+ group only. After challenge, compared to the V- inoculated group, viremia was 100-fold lower at 10 days post-infection in A-V+ whereas viremia was not significantly reduced in A+V+ piglets. A lower transmission rate was estimated for the A-V+ group: 0.15 [0.07−0.29] versus 0.44 [0.18−1.76] and 0.32 [0.14−0.68] for the A+V+ and V- groups, respectively.

Investigations about the low vaccine strain detection after the first vaccination suggested a relationship between IFNa levels and vaccine strain detection in A-V+ piglets. We showed that MDNAs impair vaccine efficacy against PRRSv both in inoculated and contact piglets, probably by reducing vaccine replication. IFNa may also interfere with PRRSv vaccination. These new data could help improving vaccination protocols.

Practical benefit

As previously shown, investigating interference between MDA and vaccine is not a new question for animal pathogens. But, to date no investigations have been conducted to explore this potential interference for PRRS MLV and the consequences in terms of efficacy reduction. Consider MDA interference with PRRS MLV will allow vaccine producers and swine practitioners to define updated vaccine protocols able to circumvent MDA interference.

Further reading

↗ *Maternally-derived neutralizing antibodies reduce vaccine efficacy against porcine reproductive and respiratory syndrome virus infection.*

2014

Award-winning projects:

➡ **Investigation of the duration of viremia and protection after simultaneously vaccination with PRRS MLV against both PRRSv type 1 and type 2**

Charlotte Sonne Kristensen. The Royal Veterinary and Agricultural University, Denmark

➡ **Interference of Maternally Derived Antibodies with PRRS vaccine in piglets**

Nicolas Rose. Anses, France

➡ **Determination of the frequency of animals with broadly cross-reactive neutralizing antibodies in the sow population**

Cinta Prieto. Universidad Complutense de Madrid, Spain

2014

Charlotte Sonne Kristensen.
The Royal Veterinary and Agricultural
University, Denmark

Investigation of the duration of viremia and protection after simultaneously vaccination with PRRS MLV against both PRRSv type 1 and type 2

Study objective

The objective of this study is to clarify a) if simultaneously vaccinated with PRRS MLV vaccines against type 1 and type 2 results in prolonged duration of PRRSv viremia b) the effect of protection against both PRRSv types after simultaneously vaccinated with PRRS MLV vaccines against type 1 and type 2.

Study design

In a laboratory challenge study, in total 66 four-weeks old PRRS naïve pigs were vaccinated with either type 1 (VAC-T1, Porcilis PRRS), type 2 (VAC-T2, Ingelvac PRRS MLV), simultaneously with both vaccines (VAC-T1/T2)or left un-vaccinated as controls. Sixty-two days later the pigs were challenged with either PRRS 1 subtype 1, PRRS 1 subtype 2 and PRRS type 2. Pigs were euthanized 2 weeks later.

Results

Results on virologic parameter following vaccination:

- PRRSv MLV vaccine was detected in serum following vaccination for 42 (VAC-T1/T2) and 62 days (VAC-T1 and VAC-T2).
- Overall, the level of virus RNA in serum of VACT1T2 pigs was not different from the VAC-T2 pigs.

Results on clinical parameter after challenge:

None of the challenges resulted in significant clinical or gross pathological lesions.

Results on serology following challenge:

After challenge with PRRSv-1, the level of virus in the NON-VAC group was in general higher than in the vac-cinated groups. Overall, development of viremia was delayed and of shorter duration in the vaccinated pigs.

PRRSv-1, subtype 1 challenge PCR – serum:
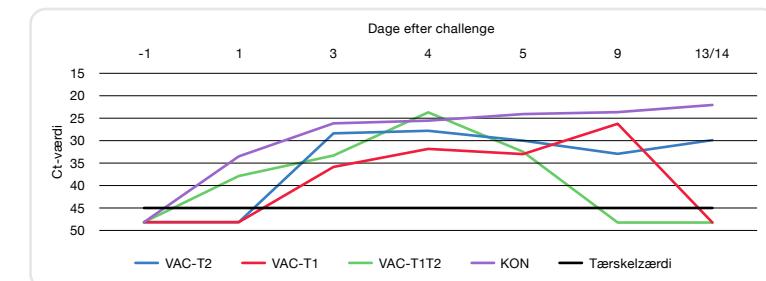
*PRRSv-1, subtype 2 challenge PCR – serum:*
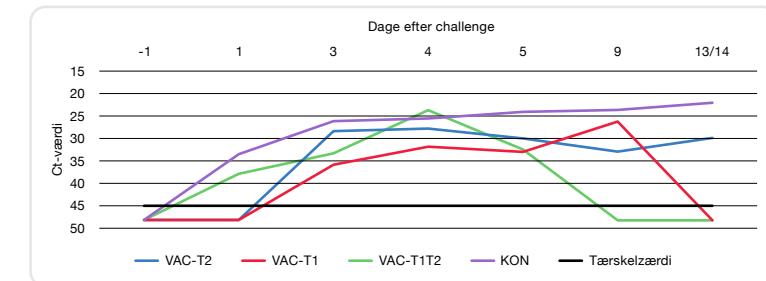
2014

Charlotte Sonne Kristensen.
The Royal Veterinary and Agricultural University, Denmark

Investigation of the duration of viremia and protection after simultaneously vaccination with PRRS MLV against both PRRSv type 1 and type 2

PRRSv-2, challenge PCR – serum:
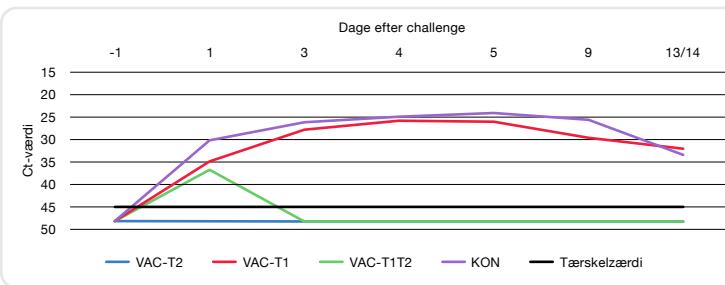
PRRSv-2 vaccinated pigs were almost completely protected from viremia following challenge with the homologous PRRSv-2 strain.

Conclusion

None of the animals experienced any adverse effect following single or simultaneous vaccination with two PRRSv MLV vaccines and the viral load and duration of viremia were comparable between the two groups. Furthermore, no differences in responses of single and dual vaccinated animals were seen after homologous and heterologous challenge.

Nicolas Rose.
Anses, France

Interference of Maternally Derived Antibodies with PRRS vaccine in piglets

Current problem

Regarding PRRS vaccines, previous studies had shown that modified live vaccine (MLV) can induce neutralizing antibodies (NA) in gilts . It was also demonstrated, that these NA can be transferred to piglets by the colostrum and can delay PRRS infection in these piglets. Even if there is no data to date for PRRS infection, we cannot exclude that immune cells transferred through the colostrum could also interfere with PRRS infection in the piglets.

Study objective

To investigate the potential interference of Maternally Derived Antibodies (MDA) with a PRRS modified live vaccination in piglets, in terms of humoral and cellular immune response.

Study design

The piglets came from a vaccinated breeding herd. Thirty piglets with a low (A) or high level (A+) of PRRSvneu-tralizing MDAs were vaccinated (V+) with a modified live vaccine at 3 weeks of age. Blood samples were collected before vaccination and then at 2, 4, 8 and 14 weeks post-vaccination (WPV). The samples were analysed to detect the vaccine viraemia (RT-PCR) and quantify the post-vaccination humoral (ELISA and virus neutralisation test) and cellular (ELISPOT IFNg) immune responses.

Results

PRRSv vaccine strain was detected in 60%, 64%, 36% and 0% of A V+ piglets 2, 4, 8 and 14 WPV respectively. No virus was detected in A+V+ piglets during the first four WPV but 32% and 6% of A+V+ piglets were PCR-positive at 8 and 14 WPV. Eighty-five percent of A-V+ piglets and 0% of A+V+ piglets seroconverted (ELISA) between 2 and 4 WPV. Neutralising antibodies appeared 4 WPV in the A-V+ piglets and 14 WPV in the A+V+ piglets. The number of PRRSv-specific IFNgsecreting cells was significantly higher in A V+ piglets at 2 and 4 WPV than in A+V+ piglets.

Conclusion

These results show that MDAs can affect both post-vaccination humoral and cellular immune responses in piglets. Further studies are required to assess the impact of MDAs on vaccine efficacy following a PRRSv challenge and its ability to reduce viral transmission.

Practical benefit

This study will help to answer questions around timing of vaccination in piglets.

Further reading

↗ *Maternally-derived antibodies (MDAs) impair piglets' humoral and cellular immune responses to vaccination against porcine reproductive and respiratory syndrome (PRRS).*

Determination of the frequency of animals with broadly cross-reactive neutralizing antibodies in the sow population

Cinta Prieto.
Universidad Complutense de Madrid, Spain

Current problem

Although the components of the immune response responsible for protection have not been definitively identified, it has been demonstrated that neutralizing antibodies might play a role in protection, at least against reinfections. Gaining better knowledge on these cross reactive neutralizing Antibodies will help to modify PRRS control approaches.

Study objective

1 To determine the frequency of elite neutralizers among the sow population and the existence of differences between farms.

2 To determine whether the acclimatization system followed in different farms might have an influence in the proportion the breeding population developing crossly reactive NA.

3 To determine whether the age of the sows has any influence in the frequency of elite neutralizers and vaccinated and unvaccinated farms.

4 To determine whether genetic has any influence in the frequency of elite responders.

Practical benefit

The results of this study will allow knowing the frequency of elite responders and, more importantly, the factors that can influence their proportion. If management or genetic factors that influence the ability of sows to develop broadly NAs are identified, new management procedures or genetic programs might be developed in the future to improve the level of protection of the herd population. Besides, if the value of NAs against reinfections can be definitively proven, relatively simple assays could be developed to determine the level of protection that can be expected in a population, using a selected panel of viruses in SN assays. In addition, this project could contribute to a bigger research project in which the location of neutralizing epitopes responsible for cross-neutralization is the main aim. If this objective is achieved, new and more easily applied diagnostic techniques could be applied to predict protection. Finally, if the haplotype of good responders can be defined, this information will be useful for future selection programs in breeding companies.

Further reading

↗ *The Ability of Porcine Reproductive and Respiratory Syndrome Virus Isolates to Induce Broadly Reactive Neutralizing Antibodies Correlates With In Vivo Protection.*

Boehringer Ingelheim