JOURNAL OF SWINE HEALTH SPRODUCTION

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Inquisitive Iowa pig

Photo courtesy of Sue Schulteis

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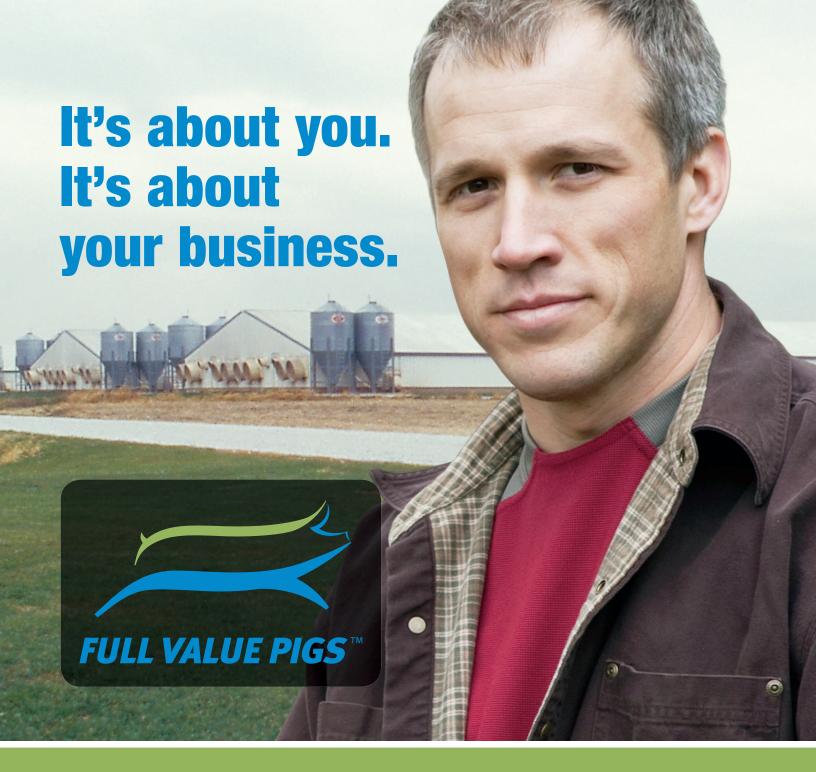
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"I have chosen veterinary medicine as a career because I enjoy working with people, animals, and science."

quoted from Why do you do what you do, page 125



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Integrity, intensity, professionalism

The 2015 AASV Annual Meeting was held in Orlando, Florida, February 28th to March 3rd. Once again this annual meeting was a valuable experience for all who attended. No doubt all veterinarians were able to take home good scientific knowledge as well as practical information to use when returning to their professional lives. For me, this annual meeting was a unique experience. As chairman of the program committee, instead of being in the audience looking on, I was on the inside looking out. I was very pleased. From the podium, I could see the record crowd consisting of seasoned veteran practitioners, many international veterinarians, young up-and-coming veterinarians, and once again, many, many students.

The experience of our association staff (Tom, Sue, Harry, and Dave), along with their assistants, made the event flow smoothly and seamlessly. The hotel venue was an excellent choice, and the hotel staff was accessible and accommodating. Orlando is a great place for a meeting.

I want to thank our speakers for the quality of their presentations, especially Dr Greg Stevenson for the Howard Dunne lecture and Dr Scanlon Daniels for the Alex Hogg lecture. Both great messages addressed our theme "Beyond Our Oath: Integrity, Intensity, Professionalism." I have reviewed both, and found more gems of knowledge that I will share with you in this letter. I

was impressed by how well all sessions drew a crowd and how evenly the attendance was spread among concurrent sessions. All provided excellent technical information. The quality of the workshops on Saturday afternoon and Sunday morning offered some difficult choices for many of us.

I also want to thank our program committee for volunteering their time and effort to put together our program. Many of them were chairpersons of a session. They were Matt Anderson, Butch Baker, Andrew Bents, Mike Brumm, George Charbonneau, Mitch Christensen, Scanlon Daniels, Monte Fuhrman, Jeff Harker, Megan Inskeep, Karen Lehe, Michelle Michalak, Chris Rademacher, Alex Ramirez, Craig Rowles, Adam Schelkopf, Michelle Sprague, Scott Stehlik, Jennifer Stevens, Matt Turner, John Waddell, Todd Williams, and Nate Winkelman. Thank you, ladies and gentlemen, for your collective time and expertise compiling a great program!

"As we maintain our integrity and professionalism, we will raise the value of swine veterinarians to the pork industry and cultivate the respect of consumers of pork."

Some highlights from our two keynote lectures: Greg Stevenson in his Howard Dunne lecture focused on integrity as a virtue in our lives and how essentially important it is to practice integrity in our veterinary profession. He cited the importance of integrity with respect to antibiotic usage, as well as quality of scientific study. Dr Stevenson emphasized the importance of healthy, constructive criticism of scientific studies as a civilized process that should not be viewed as "critical" toward an individual or entity. He said "Quality of science includes both civility and criticism." Greg is a great role model for this and has one of the best scientific minds in our profession. He also stressed the importance of identifying and disclosing conflicts of interest within our profession. One of his key observations: "The reduction in the number of decision makers combined with increased numbers of pigs

ambined with increased numbers of pigs and amounts of money involved in single decisions has changed the competitive landscape and put increased pressure on the professional integrity of swine veterinarians in their many roles." Greg and I agree. I believe we as veterinarians can withstand the pressures we face. As we maintain our integrity and professionalism, we will raise the value of swine veterinarians to the pork industry and cultivate the respect of consumers of pork.

Scanlon Daniels offered some real "gems" of advice in his Alex Hogg lecture "Influence and Advocacy: Opportunities for Swine Veterinarians." Dr Daniels is a private-practice veterinarian respected and admired by many of his colleagues. He has experience working for corporate hog production and has applied his experience to private practice. He emphasized the value of developing and maintaining relationships with the people you seek to influence. He said "For me it became clear that I had more influence with farms where I had a stronger relationship with the people." Influencing farms toward the implementation of new technology, Dr Daniels said "If science is the engine driving change, then relationships are where the rubber meets the road." Does AASV have a role in advocacy? He believes AASV as an organization "needs to assertively and proactively address societal concerns" for us to have a strong role of influence. To be taken seriously we must "continue our strengths and engage society." The new emphasis should be on "engagement." Scanlon is still a young veterinarian with wisdom beyond his years. I expect he will have longlasting influence within AASV.

There were numerous other great technical presentations that I will review in my next letter. Reference to porcine epidemic diarrhea virus (PEDV) was common throughout many of them: PEDV could have been the theme for the meeting. Fortunately, PEDV virus has been uncommon this winter, much to the relief of producers and veterinarians. Here I will close by offering credit to AASV swine veterinarians in the field and in our laboratories for putting science into practice so quickly through integrity, intensity, and professionalism.

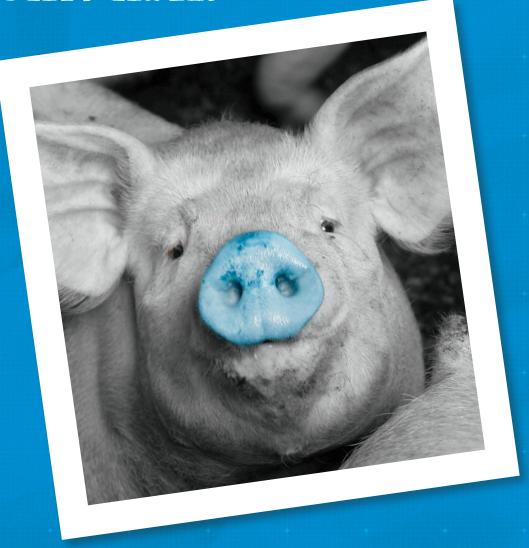
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Maximize your reading - Part 3

This is the third part of my series on maximizing your reading. In parts 1 and 2, I shared some of the strategies that I use when tackling my scientific reading.^{1,2} My intent with this series is that hopefully you will take home a few pointers to help you get the most out of your reading. A discussion on critiquing and reading the scientific literature would not be complete without some time dedicated to sample size consideration. As a researcher, I find that critiquing the way others justify sample size, as well as their sampling techniques, helps bring awareness to how I make these decisions in my own work. The sample size used in any work presented in a paper should be clear and justified. The study genre will greatly impact the sample size potential, eg, a case report may involve one animal or one farm, and a randomized controlled clinical trial may involve hundreds of animals or more or less. Many computer software programs are available for determining sample size, but there are other considerations when determining sample size, ie, a small sample size of animals may have an impact on the power of the statistical analysis which can minimize the strength of the study, and a large sample size may be associated with cost limitations for the study.³ Additionally, a study that uses many subjects is not necessarily "better" if fewer subjects would have supported and answered the scientific

question. A good scientific writer should include in the methods section a discussion on the strengths and weaknesses of the sample size used in the study and subsequently how that impacts the analysis and interpretation of the data. A good critical reviewer of a paper should also look for this discussion and take that into consideration as well when formulating his or her own conclusions.

"As you can see, all the sections of a paper build the framework for drawing conclusions from the work presented."

The statistics section is often the most difficult to critique for many readers. The task of explaining how to understand statistics in an editorial is overwhelming for me as well. I suggest purchasing a statistics text book – particularly one written for veterinary medicine – to have as a quick reference and to help guide you or to provide a quick refresher when a statistical method or test seems unfamiliar.

The results section of the paper typically follows the materials and methods. I am usually guilty of quickly skimming results if it is in text format and I constantly try to discipline myself to read this section more slowly. But I do look at the tables and figures in detail. I always keep in mind when reading a table or figure that the data is often presented as the authors' interpretation. For example, the paper may state something like "the data in Figure X show that the average daily gain of the pigs in Group B was better than in Group A". This is likely the case, but I look carefully to see if the data actually show what the authors say the data show. Of course, I cannot do this as a critical reader if I do not fully understand the methods and their limitations. As you can see, all the sections of a paper build the framework for drawing conclusions from the work presented.

The discussion section comes next. The author is expected to do a good job of examining and explaining how the work has advanced our knowledge, any new insights provided, or future research directions. Issues or controversies raised by the findings should be discussed in this section

and ideally resolved or supported with other literature.

In the Journal of Swine Health and Production (JSHAP), an implications section comes at the end of the manuscript. This is a bulleted list of take-home information for readers and should contain the practical application of the results. This section is often critiqued by reviewers as being unnecessary and not appropriate in scientific reporting. This is a difficult section for authors as well, as the temptation is to just re-iterate the results. The intent of this bulleted list is to provide a quick source of information for busy practitioners, and I think it is appropriate for an applied journal such as JSHAP. While this bulleted list is quick, easy, and often informative to all of us with busy schedules, I recommend a full review and critique of the entire paper by all readers.

A scientific paper would not be complete without references and acknowledgements. While this is seemingly obvious, references should be relevant, current, and presented neatly. Acknowledgements are important for readers to consider as well. It is essential to recognize those who participated and financially contributed to the research, as without this there would be no advancement of our knowledge.

I certainly have not covered every aspect of how to maximize your reading, but I hope this short series has provided you with some new information, reminded you of forgotten information, or inspired you to catch up on some reading.

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Terri O'Sullivan, DVM, PhD Executive Editor





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Why do you do what you do?

Why I do what I do

When Dr Burkgren offered the unexpected opportunity to write this column, an early thought was that I had previously answered this question and will be able to pull the answer out of my files. I soon discovered that the first and only time that anyone has asked a similar question and requested a written answer from me was a short....29 years ago!

The specific question at that time in December 1985 was "Why Did I Select Veterinary Medicine as a Career?" It was a part of the application to the University Of Missouri College of Veterinary Medicine. Each applicant answered the question by writing an essay and later discussing that essay and a wide range of other topics in an interview with the admissions committee. It was a great open-ended question that stopped students in their tracks and fostered the same introspection as when the senior college student asked Dr Burkgren, "Why do you do what you do?"

I was able to locate my original essay and compared what I dreamed would be rewarding in this profession to what I actually find rewarding as a swine veterinarian 25 years following graduation. The opening sentence provided a solid reminder of what drew me into this profession: "I have chosen veterinary

medicine as a career because I enjoy working with people, animals, and science."

Thinking of the current day, one of the best parts of being a swine veterinarian truly is working with a wide variety of people. I have the opportunity daily to interact and collaborate with many talented swine producers, co-workers, and veterinarians across the United States and Canada as part of my employment within an agricultural cooperative's pig contracting business. I am privileged to be part of great teams that have thrived for many years that include coworkers, farm owners, farm employees, and veterinary colleagues. The dynamic nature of the swine industry continually creates the need to meet, interact, and team up with new people to accomplish goals, solve problems, and share ideas.

It is a rewarding part of my job to experience the success of farms in terms of swine health and the personal growth of people, and as measured in business terms. Those farms, people, and animals play a vital role in global society by producing safe, nutritious, and affordable food for a world population that's growing by 200,000 people every day. Being a part of their effort makes it easy to find relevance and importance in serving society as a swine veterinarian. It's engaging to be on the worldwide team of agriculture that currently shoulders the goals and challenges of producing food for 7.3 billion people.

My interest in becoming a veterinarian was launched during early grade school by working with pigs on our family farm. My first experience with veterinarians was watching them on farm calls assisting our family with mortality caused by diseases such as Escherichia coli and erysipelas. It was fascinating to watch a veterinarian examine live pigs or perform a post mortem exam, reach a diagnosis, and recommend treatment in a matter of minutes. By the following day, the pigs were responding well to treatment and preventative measures were started for the future. That was (and is) applied science at its very best! I continue to enjoy and appreciate the rapid response that swine

veterinary medicine provides in defining and solving problems for animals and people. That rapid response occurs at the farm level, the diagnostic laboratories, and many other places in the middle.

"I have chosen veterinary medicine as a career because I enjoy working with people, animals, and science."

There was one pending reward of becoming a swine veterinarian that was completely unanticipated by me as an undergraduate. That reward is membership in the AASV. It's exceptionally motivating to be part of a professional association in which members selflessly educate and challenge each other in the areas of veterinary practice, science, and integrity. The members and the AASV staff truly create a professional home that encourages all of us to become better at doing what we do.

In conclusion, I find that the final sentence of my essay was an accurate predictor of the reality of being a swine veterinarian: "I believe a career in veterinary medicine will be enjoyable, challenging, and rewarding."

Being a swine veterinarian is all of that and a whole lot more! That's why I do what I do!

Bill Starke, DVM Pig Sourcing Team Manager Purina Animal Nutrition LLC – A Land O'Lakes, Inc. company





ORIGINAL RESEARCH

Monitoring influenza A virus infection in pigs by using a competitive enzyme-linked immunosorbent assay to detect virus antibodies in pen-based oral-fluid specimens

Katrin Strutzberg-Minder, MSc, DSc; Jan Boehmer, DVM; Sebastian Fischer, DVM; Matthias Homuth, DVM; Oliver Gomez-Duran, DVM, PhD, MRCVS, Certificate in Pig Medicine, Diplomate ECPHM; Gudrun Finger, veterinarian; Marika Genzow, DVM, Diplomate ECPHM

Summary

Objective: To investigate monitoring an influenza A virus (IAV) infection in a finishing pig herd by testing pen-based oral fluids for antibodies against the virus using a commercial enzyme-linked immunosorbent assay (ELISA) kit.

Materials and methods: Oral fluids were collected weekly from pigs 12 to 24 or 22 weeks of age in four pens (approximately 25 pigs per pen) in two consecutive batches. Serum samples were also collected from two randomly selected pigs in each pen at 12, 16, and 20 weeks of age in both batches and at 24 weeks in Batch 1 only. Oral-fluid and

serum samples were tested for antibodies against IAV by a commercial competitive ELISA test kit. Oral fluids were also tested for IAV by reverse transcription-polymerase chain reaction (RT-PCR).

Results: One week after initial detection of IAV in oral-fluid samples by RT-PCR, antibodies against the virus were detected in oral fluids as well as in serum samples. Oral fluids continued to test positive for antibodies 4 to 7 weeks after initial detection of virus, but with a decreasing trend in the amounts of virus antibodies detected by ELISA. All samples in Batch 1 tested negative after 9 weeks.

Implications: The longitudinal profile of antibodies against IAV detected in oral fluids promises to be a useful tool for monitoring IAV infection in a pig population. A commercial competitive ELISA test kit could easily be adapted for oral fluids by modifying dilution of the specimen.

Keywords: swine, oral fluid, antibody, influenza A virus, monitoring

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Resumen - Monitoreo de la infección del virus A de influenza en cerdos utilizando la prueba de ensayo por inmunoabsorción ligado a enzimas competitiva para detectar anticuerpos contra el virus en muestras de fluidos orales por corral

Objetivo: Investigar monitoreando una infección del virus A de influenza (IAV por sus siglas en inglés) en cerdos de finalización por medio del análisis de fluidos orales en corral en busca de anticuerpos contra el virus utilizando una unidad de análisis comercial del ensayo por inmunoabsorción ligado a enzimas (ELISA por sus siglas en inglés).

Materiales y métodos: Semanalmente se recolectaron fluidos orales de cerdos de 12 a

24 o 22 semanas de edad en cuatro corrales (aproximadamente 25 cerdos por corral) en dos grupos consecutivos. También se recolectaron muestras de suero de dos cerdos seleccionados al azar en cada corral a las 12, 16, y 20 semanas de edad en ambos grupos y a las 24 semanas de edad solamente del Grupo 1. Se analizaron las muestras de suero y fluidos orales en busca de anticuerpos contra el IAV por medio de una unidad comercial de ELISA competitiva. También se analizaron fluidos orales en busca de IAV por medio de la reacción en cadena de la polimerasa de transcriptasa inversa (RT-PCR por sus siglas en inglés).

Resultados: Una semana después de la detección inicial de IAV en las muestras

de fluidos orales por medio de la RT-PCR, se detectaron anticuerpos contra el virus en fluidos orales y en las muestras de suero. Los fluidos orales continuaron dando resultados positivos a los anticuerpos 4 a 7 semanas después de la detección inicial del virus, pero con una tendencia descendente en la cantidad de anticuerpos virales detectados por ELISA. Todas las muestras en el Grupo 1 resultaron negativas después de 9 semanas.

Implicaciones: El perfil longitudinal de anticuerpos contra el IAV detectados en fluidos orales aparenta ser una herramienta útil para el monitoreo de la infección de IAV en una población porcina. Una unidad de prueba comercial de ELISA competitiva podría ser adaptada fácilmente para fluidos orales, modificando la dilución del espécimen.

Résumé - Surveillance de l'infection par le virus influenza A chez des porcs à l'aide d'une épreuve immunoenzymatique compétitive afin de détecter au niveau des enclos des anticorps contre le virus dans des échantillons de fluides oraux

Objectif: Étudier la surveillance d'une infection par le virus influenza A (VIA) dans un troupeau de porcs en finition en testant des

KSM, JB, SF, MH: IVD Innovative Veterinary Diagnostics (IVD GmbH), Hannover, Germany.

OGD, MG: Boehringer Ingelheim Animal Health, Ingelheim am Rhein, Germany.

GF: Veterinary Clinic Lindhaus, Schöppingen, Germany.

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This article is available online at http://www.aasv.org/shap.html.

Strutzberg-Minder K, Boehmer J, Fischer S, et al. Monitoring influenza A virus infection in pigs by using a competitive enzyme-linked immunosorbent assay to detect virus antibodies in pen-based oral-fluid specimens. *J Swine Health Prod.* 2015;23(3):126–131.

fluides oraux prélevés au niveau des enclos pour des anticorps dirigés contre le virus à l'aide d'une épreuve immunoenzymatique (ELISA) vendue commercialement.

Matériels et méthodes: Des fluides oraux furent prélevés sur une base hebdomadaire chez des porcs âgés de 12 à 24 ou de 22 semaines dans quatre enclos (environ 25 porcs par enclos) lors de deux lots consécutifs. Des échantillons de sérums furent également prélevés de deux porcs choisis de manière aléatoire dans chacun des enclos à 12, 16, et 20 semaines d'âge dans les deux lots, et à 24 semaines dans le Lot 1 uniquement. Les échantillons de fluides oraux et de sérum furent testés pour la présence d'anticorps contre le VIA au moyen d'une trousse commerciale d'un test ELISA compétitif. Les fluides oraux furent également testés pour le VIA au moyen d'une réaction d'amplification en chaîne en utilisant la transcriptase réverse (RT-PCR).

Résultats: Une semaine après la détection initiale par RT-PCR de VIA dans des échantillons de fluides oraux, des anticorps dirigés contre le virus furent détectés dans les fluides oraux ainsi que dans les échantillons de sérum. Les fluides oraux étaient toujours positifs pour la présence d'anticorps 4 à 7 semaines après la détection initiale du virus, mais avec tendance à la baisse dans les quantités d'anticorps contre le virus détectées par ELISA. Tous les échantillons du Lot 1 étaient négatifs après 9 semaines.

Implications: Le profil longitudinal de détection d'anticorps contre VIA dans des fluides oraux promet de s'avérer un outil utile pour surveiller l'infection par le VIA dans une population de porcs. Une trousse commerciale d'un test ELISA compétitif a pu facilement être adaptée pour des fluides oraux en modifiant la dilution du spécimen.

onitoring and surveillance of infectious diseases in swine populations is a key component in prevention or control of clinical losses, but is often limited by the cost and inconvenience of collecting individual samples. Analysis of pen-based oral-fluid samples has proven to be an efficient method for monitoring and surveillance of various infectious diseases in swine populations.¹ It has been shown that oral fluid is an appropriate specimen for direct detection of important viruses in pig production, eg, porcine reproductive and respiratory syndrome virus (PRRSV),² porcine circovirus type 2 (PCV2),³ and influenza

A virus (IAV).^{4,5} However, detection of an infectious agent is not diagnostic proof of either an infection or an infectious disease. For this reason, detection of antibodies against an infectious agent as an immunological response to the agent is of additional usefulness in monitoring and surveillance. Antibodies against various viral agents, eg, PRRSV,⁶ PCV2,³ and IAV,⁷ have been detected in oral fluids.

Influenza is an important component of the porcine respiratory disease complex (PRDC) and a pathogen with a major economic impact in commercial swine populations. It has been shown that a commercial blocking enzyme-linked immunosorbent assay (ELISA) designed to detect influenza A nucleoprotein (NP) antibodies in avian serum accurately detects NP antibodies in swine serum.8 Furthermore, oral-fluid samples from pigs infected with influenza virus have been shown to contain detectable concentrations of antibodies against the NP of the virus from 7 to 42 days post inoculation, and detection of these antibodies is possible using a commercial blocking ELISA.7 In a longitudinal study in two pig farms,⁹ the dynamics of IAV infection determined by polymerase chain reaction (PCR) analysis of nasal swabs differed from those determined by testing serum using a commercial ELISA and by a hemagglutination inhibition (HI) test. Therefore, the objective of this study was to investigate the practical feasibility of monitoring IAV infection dynamics in a commercial pig herd by testing for anti-NP antibodies in pen-based oral-fluid samples. For that purpose, results of testing serum samples, still routinely used for antibody PRDC monitoring in Germany, 10 were compared to results of testing oral fluids.

Materials and methods

The trial involved no intervention in the animals or medications requiring notification or exemption. The farm complies with German production standards and animal welfare regulations.

Experimental design

The suitability of pen-based oral fluids for monitoring IAV infection in a conventional finishing pig herd was compared to the usual monitoring method, ie, testing serum samples. Status of influenza infection was monitored by testing oral fluids for IAV by a conventional reverse transcription PCR (RT-PCR).

Pig herd

The trial was conducted from June 2013 until January 2014 in two successive batches of finishers in a commercial herd in northwest Germany. Each batch consisted of approximately 100 pigs housed in one room with four pens, with approximately 25 pigs per pen. The herd had a history of circulating PRRSV, IAV, and Mycoplasma *hyopneumoniae*. This was a closed system in which the sow herd had been vaccinated with Ingelvac PRRSV MLV (Boehringer Ingelheim Vetmedica Inc, St Joseph, Missouri) every 3 months. The sows were clinically healthy when the piglets used in the study were produced. Piglets were weaned at day 27 or 28 of life and moved to the finishing barn at 10 to 12 weeks of age.

Porcine samples

Oral fluids were collected once a week from each of the four pens, starting when the pigs were 12 weeks old and continuing until they were 24 weeks old for Batch 1 and 22 weeks old for Batch 2. For collection of samples from each pen, one rope was used, which remained in the pen 10 to 15 minutes. Samples were collected in the morning prior to feeding, then chilled and shipped on ice within 24 hours to the IVD GmbH laboratory, Hannover, Germany. Samples were collected using the Swine Oral Fluids Collection Kit (ITL Animal Healthcare, Reston, Virginia). For comparison, serum samples were also collected from two pigs from each pen at 12, 16, and 20 weeks for both batches, and at 24 weeks for Batch 1 only. Pigs for serum collection were randomly selected by drawing two animal identification numbers from a container. Samples were tested on the day they were received by the laboratory and stored at 4°C until all data had been entered in the laboratory information system (Ticono-LC [LabControl]; Ticono GmbH, Hannover, Germany).

Influenza A virus ELISA

All oral-fluid and serum samples were tested for antibodies against the NP of IAV using a commercially available competitive IAV ELISA for serum samples from poultry and swine (ID Screen Influenza A Antibody Competition; IDvet, Grabels, France; officially registered in Germany). Oral-fluid samples were diluted 1:2 using the dilution buffer provided with the test kit, and serum samples were diluted 1:40 as described in the test-kit instructions. For all other

procedures, the manufacturer's instructions were followed exactly. The ELISA results were reported as a ratio percentage (S:N%) of the optical density (OD) of the sample (S) to the OD of the negative control (N). For porcine serum, S:N% \leq 45% are classified positive; values > 45% and < 50% are classified as questionable; and values \geq 50% are classified as negative.

Influenza A virus RT-PCR

Nucleic acid was extracted from oral-fluid samples using an RNA-DNA isolation kit (MagMAX Pathogen RNA/DNA Kit; Life Technologies GmbH, Darmstadt, Germany) and an automated nucleic acid isolation processor (MagMAX Express-96 Magnetic Particle Processor; Life Technologies GmbH) that used magnetic bead technology. A total of 450 µL of lysis buffer from the kit was added to $300 \,\mu\text{L}$ of each oral-fluid sample, suspensions were vortexed for 3 minutes, and cell debris was removed by centrifugation for 2 minutes at 16,000g. Then, 600 µL of each supernatant was transferred on a 96-well plate into the processor, and nucleic acid isolation was performed according the manufacturer's protocol and instructions. Nucleic acid extracts were analyzed by RT-PCR as previously described, with the following modifications.¹¹ The assay was performed as a two-step RT-PCR using M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant, and random hexamers (Promega GmbH, Mannheim, Germany) for firststrand cDNA synthesis, 4 µL of first-strand cDNA reaction for second-strand cDNA synthesis, and PCR amplification according to the manufacturer's recommendations. The PCR was performed in a Mastercycler (Eppendorf AG, Hamburg, Germany) with the following cycling conditions: 95°C for 2 minutes, followed by 40 cycles of 95°C for 30 seconds, 57°C for 30 seconds, and 72°C for 30 seconds, followed by 72°C for 2 minutes, and 15°C until each reaction was analyzed by agarose gel electrophoresis and ethidium bromide staining. Every sample was monitored for inhibition by dividing the PCR mix and adding IAV cDNA to one of the halves; each run of RT and PCR was monitored by a no-template control and an IAV-positive control (RNA prepared from supernatant of an IAV cell culture). The limit of detection in oral fluids spiked with IAV strains of each of the subtypes H3N2, H1N1, and H1N2 was at least 10⁴ median tissue culture infective doses per mL.

Results

Influenza A virus RT-PCR

The IAV was detected in one pen-based oral-fluid sample from each batch when the pigs were 15 weeks of age (Tables 1 and 2). All other samples collected (51 of 52 from Batch 1 and 43 of 44 for Batch 2) were negative by RT-PCR.

Influenza A virus ELISA

Antibodies against IAV were detected both in oral fluids and serum (Figures 1 and 2) for both batches starting at 16 weeks of age. This was 1 week after the first detection of IAV RNA by RT-PCR in the oral fluids. Antibodies were no longer detectable in oral fluids in some pens starting at 20 weeks for Batch 1 and 22 weeks for Batch 2, whereas serum samples stayed positive until the last sampling date, at 24 weeks for Batch 1 and 20 weeks for Batch 2. At 12 weeks, four of eight serum samples from individual pigs of Batch 1 and six of eight serum samples from Batch 2 were ELISA-positive, but oral-fluid samples were all negative.

Two or 3 weeks after detection of IAV in the herd by oral-fluid RT-PCR (Figures 1 and 2), there was a decreasing trend in the oral-fluid ELISA results from all four pens in both batches, as demonstrated by the increasing S:N% values of the competitive ELISA. By comparison, S:N% values of the serum samples from both batches were consistently positive (mean of ELISA S:N% 4.5 to 8.4 with a maximum standard deviation of 4.0).

Discussion

While serum samples are still widely used for routine monitoring and surveillance programs in Germany, in most cases the quality of oral-fluid specimens is appropriate for direct and indirect detection of infectious agents¹⁻⁷ as long as the guidelines for collection are adhered to strictly. A pen-based oral-fluid sample is an aggregate sample from multiple animals, just as a sample from a bulk milk tank is an aggregate sample. In this case, infected animals are more likely to contribute to the sample. It has been shown for PRRSV that testing penbased oral-fluid samples greatly improved detection over single-animal testing.¹² Furthermore, this is a very convenient procedure for collecting a specimen from an animal-welfare perspective. Because of the convenience of this sampling procedure

for both humans and animals, it can be used weekly to provide very informative, infection-correlated monitoring. While bi-weekly sampling may be sufficient in terms of the cost-benefit ratio for PRDC monitoring, and therefore is recommended by multiple experts, 13 our results indicate that the sampling interval should not be longer than 2 weeks. Otherwise, there is a loss of information about the dynamics of an infection and those of the herd immunity. With bi-weekly sampling, an IAV infection is detected at the latest 2 weeks after infection, whereas it takes 4 weeks, or twice as long, with monthly sampling. Furthermore, the phase of infection can be determined within the shorter time period of 14 days instead of a month, which is helpful for implementing strategies for prevention at the right time. These aspects should be weighed against each other when conducting a monitoring program.

Simply by adapting the dilution of oral fluids to account for the lower concentration of antibodies in oral fluids than in serum, we were able to obtain meaningful and useful antibody profiles in two consecutive finishing batches using a commercial competitive ELISA to monitor the dynamics of naturally occurring IAV infection. While other investigators have already evaluated an NP-blocking ELISA for detection of NP antibodies in oral fluids from pigs infected under experimental conditions,⁷ further studies are still needed to determine whether a commercial NP-blocking ELISA can also be used for monitoring naturally occurring IAV infections in herds. Nonetheless, the results presented here are very promising. A particular advantage of modifying an NP-blocking or a competitive ELISA for a different sample matrix is the fact that this ELISA format can in principle detect and bind the antibodies of all test antigen-specific classes of immunoglobulin (Ig) to the NP protein. Differences in the Ig classes of different sample matrices therefore, in principle, play no role in the test results. On the other hand, with an indirect ELISA, only Ig classes can be detected for which the conjugates used in the ELISA are specific. In most cases, enzyme-conjugated secondary antibodies against IgG are used in conventional indirect ELISAs. Depending on their specificity, these primarily detect only the IgG that binds the test antigen. Other classes of NP protein-binding Igs, such as IgA and IgM,

Table 1: Results of a conventional reverse transcriptase-polymerase chain reaction (PCR) test to detect influenza A virus in oral fluids collected from pigs in a commercial finishing facility in Germany (Batch 1)*

Oral fluids						Age o	f pigs (v	veeks)					
(pen)	12	13	14	15	16	17	18	19	20	21	22	23	24
60A	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg
60B	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg
61A	Neg	Neg	Neg	Pos	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg
61B	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg

^{*} Oral fluids were collected once weekly from two consecutive batches of pigs in four pens (60A, 60B, 61A, and 61B; approximately 25 pigs and one rope per pen) from 12 weeks to 24 weeks of age. Blood samples were collected from two randomly selected pigs per pen. The oral-fluid samples were tested for influenza A virus by PCR and for virus NP antibodies using a commercial competitive ELISA for porcine serum samples, conducted to confirm the results of virus NP antibody detection in those oral fluids and serum samples from the same pens. All testing was conducted to investigate the usefulness of oral fluids in comparison to serum for influenza A virus monitoring in a finishing pig herd. Positive (Pos): a clearly visible band after ethidium bromide staining of the agarose gel. The lack of a visible band was interpreted as negative (Neg). A weakly colored band, not observed in this study, would have been interpreted as a weakly positive result.

NP = nucleoprotein; ELISA = enzyme-linked immunosorbent assay.

Table 2: Results of a conventional reverse transcriptase-polymerase chain reaction (PCR) test to detect influenza A virus in oral fluids collected from pigs in a commercial finishing facility in Germany (Batch 2)*

Oral fluids						Age o	f pigs (w	eeks)					
(pen)	12	13	14	15	16	17	18	19	20	21	22	23	24
60A	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	ND	ND
60B	Neg	Neg	Neg	Pos	Neg	Neg	Neg	Neg	Neg	Neg	Neg	ND	ND
61A	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	ND	ND
61B	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg	ND	ND

^{*} Study described in Table 1.

Neg = negative; Pos = positive; ND = not determined; PCR testing not performed for pigs 23 and 24 weeks of age.

will not be detected by an indirect ELISA to an extent depending on the specificity of the conjugate used. Differences in the composition of NP-specific Ig classes in different sample matrices therefore have a considerable effect on the test results of an indirect, conjugate-dependent ELISA, because the NP-binding IgG can be detected, but in principle other NP-binding Ig classes, such as IgM and IgA, cannot be detected.¹⁴ It would be very practical and cost-effective if the same ELISA kit could be used for different sample matrices simply by appropriate modifications, as has been shown for other ELISA tests. For example, swine Salmonella antibody ELISA kits have been used to detect Salmonella antibodies in porcine serum and meat juice in European monitoring programs.¹⁵ Furthermore, ELISA tests are used for detecting antibodies to Mycobacterium paratuberculosis in bovine serum or milk (Idexx MAP

Antibody Test Kit; Idexx Laboratories, Westbrook, Maine; available in the United States and Canada).¹⁶

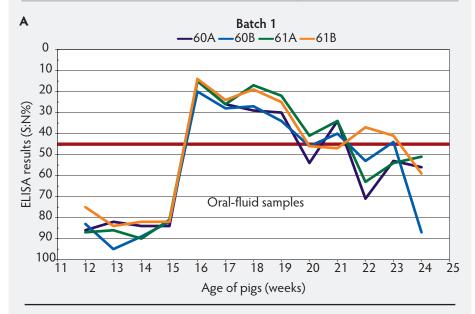
In our longitudinal study we also detected NP antibodies in both oral fluids and serum 1 week after detection of IAV in oral fluids by PCR, as did researchers in another study.⁷ But in contrast to that study, the NP antibodies we detected in oral fluids after PCR detection of IAV showed a decreasing trend within 2 to 3 weeks after detection of virus, and were no longer detectable in oral fluids in some of the pens starting at 5 weeks after exposure to IAV. One reason for these differences may be that those researchers performed an experimental infection study,7 whereas we monitored a naturally occurring IAV infection in a conventional finishing herd.

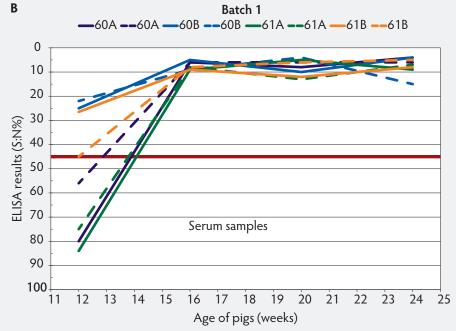
In our study, at least half of the individual serum samples tested positive for NP

antibodies 3 weeks before IAV was detected in oral fluids by PCR, while all oral fluids tested negative until IAV was detected. However, as the objective of our study was to compare the usefulness of oral fluids and serum for antibody monitoring purposes, no nasal swabs were collected, and the infectious state of the pigs for IAV at the age of 12 weeks remains ambiguous. Since the herd had a history of circulating IAV, positive serum samples at the age of 12 weeks may have been induced by an infection earlier than the beginning of our study, whereas any NP antibodies in oral fluids may have already disappeared at this time. Ultimately, this can only be clarified by further analyses.

As known from other studies, ¹⁷ maternally derived antibodies can be detected using the HI test for up to approximately 70 days. Although we did not perform the HI test for all samples in this study, it is improbable that the NP antibodies detected in serum

Figure 1: Enzyme-linked immunosorbent assay (ELISA) results for detection of antibodies against the nucleoprotein of influenza A virus (A) in oral fluids (described in Table 1) and (B) in serum samples collected monthly from two randomly selected pigs of Batch 1 in the same pens (described in Table 1). ELISA results were reported as a ratio percentage (S:N%) of the optical density (OD) of the sample (S) to the OD of the negative control (N). For porcine serum, values of S:N% ≤ 45% are classified positive (red line indicates this cut-off point); values > 45% and < 50% are classified as questionable; and values ≥ 50% are classified as negative.





by ELISA at 12 weeks of age (up to 84 days) were maternally derived, but this possibility cannot be excluded with certainty.

A shorter persistence of NP antibodies in oral fluids would be advantageous for monitoring, because detection of longer-lasting antibodies in serum might mask short periods of circulating IAV.⁹ The longitudinal profiles of

antibodies against IAV detected in pen-based oral fluids in this study were very useful for monitoring the circulation of IAV in this finishing pig herd. The efficiency and possible advantage of monitoring for IAV by antibody detection in pen-based oral fluids should be evaluated further.

Implications

- Under the conditions of this study,
 IAV infection may be detected 1 week
 after virus presence is demonstrated in
 the herd by PCR using a commercial
 competitive ELISA to detect antibodies
 against the NP of IAV in serum and
 pen-based oral fluids.
- A commercial competitive IAV ELISA designed for porcine serum samples can be used for pen-based oral fluids simply by adapting the dilution of oral fluids.
- Monitoring IAV infection dynamics in a commercial pig population is possible by detecting antibodies against the virus in pen-based oral fluids.

Acknowledgements

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Conflict of interest

None reported.

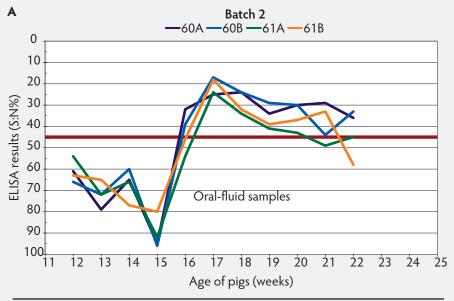
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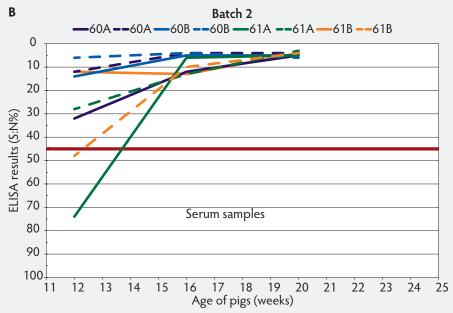
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Figure 2: Enzyme-linked immunosorbent assay (ELISA) results for detection of antibodies against the nucleoprotein of influenza A virus (A) in oral fluids (described in Table 1) and (B) in serum samples of Batch 2 in the same pens (described in Table 1). ELISA results were reported as a ratio percentage (S:N%) of the optical density (OD) of the sample (S) to the OD of the negative control (N). For porcine serum, values of S:N% \leq 45% are classified as positive (red line indicates this cut-off point); values > 45% and < 50% are classified as questionable; and values \geq 50 are classified as negative.





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BRIEF COMMUNICATION

Persistence of porcine reproductive and respiratory syndrome virus and porcine circovirus type 2 in bacterial biofilms

Mario Jacques, PhD; Daniel Grenier, PhD; Josée Labrie, MSc; Chantale Provost, PhD; Carl A. Gagnon, DVM, PhD

Summary

The aim of this pilot project was to investigate association of viruses with bacterial biofilms. Our preliminary data indicate that important viral pathogens of swine, namely, porcine reproductive and respiratory syndrome virus and porcine circovirus type 2, can associate with and persist within bacterial biofilms for several days.

Keywords: swine, porcine reproductive and respiratory syndrome virus, porcine circovirus type 2, persistence, bacterial biofilms

Received: May 8, 2014 Accepted: September 16, 2014 Resumen - Persistencia del virus del síndrome reproductivo y respiratorio porcino (PRRSV por sus siglas en inglés) y del circovirus porcino tipo 2 (PCV2 por sus siglas en inglés) en biofilms bacterianos

La meta de este proyecto piloto fue investigar la asociación de virus con biofilms bacterianos. Nuestra información preliminar indica que importantes patógenos virales porcinos, o sea, virus del síndrome reproductivo y respiratorio y circovirus porcino tipo 2, pueden asociarse y persistir dentro de los biofilms bacterianos por varios días.

Résumé - Persistance du virus du syndrome reproducteur et respiratoire porcin (VSRRP) et du circovirus porcin de type 2 (CVP2) dans des biofilms bactériens

Le but de ce projet pilote était d'examiner l'association de virus avec des biofilms bactériens. Nos données préliminaires indiquent que des virus pathogènes importants chez le porc, en particulier le virus du syndrome reproducteur et respiratoire porcin et le circovirus porcin de type 2, peuvent être associés et persister pendant plusieurs jours à l'intérieur de biofilms bactériens.

B acterial biofilms are structured clusters of bacterial cells that are enclosed in a self-produced polymer matrix and attached to a surface. ^{1,2} Biofilms protect bacteria and allow them to survive and thrive under hostile environmental conditions. Bacteria within a biofilm are usually more resistant to elimination by immune cells and to the action of antibiotics and disinfectants. The latter represents an important problem for the food industry.³

The biofilm polymer matrix might also be able to protect viruses. It has been reported⁴ that the largemouth bass virus can associate with biofilms produced by environmental strains of *Pseudomonas*, and consequently the virus is protected against certain chemical disinfectants. Moreover, biofilms in drinking-water distribution systems can

become reservoirs for pathogens, including enteric viruses.⁵ We thus hypothesized that important viral pathogens of swine can associate with bacterial biofilms and persist for long periods in the environment of swine farms. The aims of this pilot project were to investigate the association of two important viral pathogens of swine, namely, porcine reproductive and respiratory syndrome virus (PRRSV; an enveloped virus) and porcine circovirus type 2 (PCV2; a non-enveloped virus) with bacterial biofilms, and to determine whether bacterial biofilms can protect PCV2 against disinfectants.

Materials and methods

Bacterial biofilms

A standard microtiter plate assay for biofilm formation that is routinely used in our labo-

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ratory, and which involves staining biofilms with crystal violet, was performed.⁶ First, biofilms of enteric bacterial pathogens (Escherichia coli strain ECL 17608 or Salmonella Typhimurium strain ATCC 14028) and respiratory bacterial pathogens (Actinobacillus pleuropneumoniae serotype 1 strain 719 or Streptococcus suis serotype 2 strain 735 and non-typeable strain 1097925) were established in vitro following an incubation of 24 to 72 hours. The growth conditions enabling optimal biofilm formation for these bacterial strains have been already determined in our laboratory.⁷⁻⁹ In some experiments, biofilms were visualized by confocal laser scanning microscopy.⁷

Persistence of PRRSV and PCV2 in bacterial biofilms

A defined amount of a virus preparation (PRRSV genotype 2 reference strain IAF-Klop, $10^{3.0}$ median tissue culture infective doses [TCID $_{50}$] per well; or PCV2b strain FMV06-0732, $10^{4.5}$ TCID $_{50}$ per well) was added to the culture of each of the five named bacterial pathogens and incubated in a standard microtiter plate assay for bio-film formation. The persistence of the viral genome was monitored for up to 3 days in both the supernatant (ie, the liquid phase above the biofilm) and the biofilm attached at the bottom of the well. Virus-specific quantitative polymerase chain reaction

(qPCR) and reverse-transcriptase qPCR (RT-qPCR) assays were performed according to standardized protocols used by the Diagnostic Services (Faculté de médecine vétérinaire, Université de Montréal), namely, an in-house assay for PCV210 and a commercial kit for PRRSV (EZ-PRRSV MPX 4.0; Tetracore, Rockville, Maryland). The qPCR and RT-qPCR results were expressed in TCID₅₀ per mL after arithmetically comparing them to standard curves previously established with infectious PRRSV and PCV2 titrated in cell cultures. Since a positive qPCR result does not necessarily correlate with infectious potential, viral infectious titers were also determined using specific permissive cell culture models: MARC-145 for PRRSV¹¹ and NPTr¹² for PCV2 (a new cell line permissive to PCV2; unpublished data). Bacterial cells were removed by filtration on a 0.22-µm pore size membrane (UFC30GVOS; EMD Millipore, Mississauga, Ontario, Canada) before titration. The amount of infectious virus was calculated from a 96-well microplate of infected cells by the Kärber method, and the results were expressed in TCID₅₀ per mL.¹⁰ The survival of the virus within a biofilm was arithmetically compared to the survival of an equal amount of virus in a microtiter plate well in the absence of a biofilm.

PCV2 susceptibility to disinfectants in the presence of bacterial biofilms

In addition to evaluating viral persistence within bacterial biofilms, this study also determined whether bacterial biofilms can protect PCV2 (a non-enveloped virus known to be more resistant than PRRSV, an enveloped virus) against disinfectants. Porcine circovirus type 2 virions, in the presence or absence of A pleuropneumoniae biofilms, were exposed for 30 minutes to several classes of disinfectants routinely used on farms at the concentrations recommended by the manufacturers (1% acid peroxygen; Virkon, Vétoquinol, Lavaltrie, Quebec, Canada; and 1% quaternary ammonium-glutaraldehyde; Aseptol 2000, S.E.C. Repro Inc, Ange-Gardien, Quebec, Canada). A virus-specific qPCR assay could not be used in these experiments, since a positive qPCR does not correlate with infectious potential. The infectious viral titers were thus determined using the appropriate cell line as described. To ensure that residual disinfectant did not interfere with the assay, excess disinfectant was removed by ultracentrifugation at 100,000g for 1 hour, and the

virus pellet was resuspended in water to the initial volume. The viability of the bacterial cells within the biofilm was also evaluated after exposure to the disinfectants using the CellTiter-Blue cell viability assay (Promega, Madison, Wisconsin).

Results

Persistence of PRRSV and PCV2 in bacterial biofilms

An example of a bacterial biofilm formation is shown in Figure 1. Results of monitoring the presence of PRRSV for up to 3 days using a virus-specific qPCR assay are presented in Table 1. A small proportion of the viral inoculum persisted in the biofilms for the duration of the experiment; this amount was considered too small, however, to attempt quantification of infectious viruses by titration on the MARC-145 cell line. For example, the amount of PRRSV recovered from the *E coli* biofilm was 16 to 44 TCID₅₀ per mL for all time points tested, compared to a

much greater amount in positive control wells $(4831 \text{ to } 5880 \text{ TCID}_{50} \text{ per mL})$. Results of monitoring for the presence of PCV2b using the qPCR assay showed, again, that a portion of the viral inoculum persisted in the biofilms for the duration of the experiment (Table 2). These viruses were infectious when inoculated onto NPTr cells (data not shown).

PCV2 susceptibility to disinfectants in the presence of bacterial biofilms

In the second set of experiments, a defined amount of PCV2b virus strain FMV06-0732 was added to a culture of *A pleuropneumoniae*, assayed for biofilm formation, and subsequently treated with disinfectants. Results showed that PCV2b titers were lower in the presence of each disinfectant than in the negative control (ie, wells without disinfectants) and that the efficacy of the disinfectants against PCV2b was only slightly lower in the presence of the *A pleuropneumoniae* biofilm than in the control (Table 3).

Figure 1: Confocal laser scanning microscopic image of a biofilm of *Actinobacillus pleuropneumoniae*, an important bacterial swine pathogen. This is a top view of a biofilm formed at the bottom of a microtiter plate well after an incubation of 5 hours. The biofilm was stained with wheat germ agglutinin (WGA)-Oregon Green 488, a lectin that binds to poly-N-acetylglucosamine in the biofilm matrix.

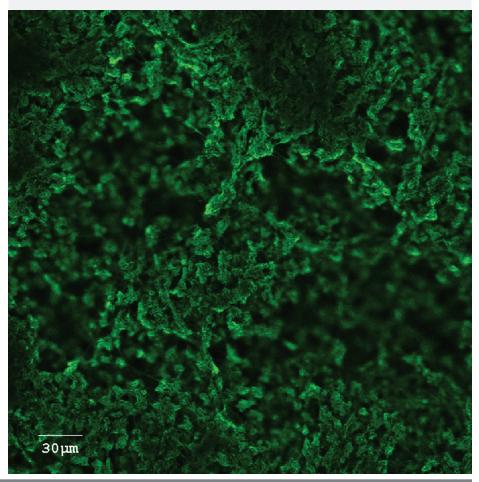


Table 1: Results of PCR testing for PRRSV in biofilms of *Actinobacillus pleuropneumoniae, Escherichia coli*, *Salmonella*, and *Streptococcus suis*

TCID₅₀ of PRRSV/mL

		1 512 30 51 1 1115 1 7 1112		
		24 hours	48 hours	72 hours
A pleuropneumoniae	Supernatant	359	2109	858
	Biofilm	27	14	4
	Positive control†	2427	4024	685
	Negative control†	0	0	0
E coli	Supernatant	5758	4409	3214
	Biofilm	16	14	44
	Positive control†	4831	5292	5880
	Negative control†	0	0	0
Salmonella	Supernatant	252	344	219
	Biofilm	21	28	53
	Positive control†	1464	3329	3622
	Negative control†	0	0	0
S suis	Supernatant	2487	2828	2638
735‡	Biofilm	106	67	276
	Positive control†	5035	3713	3031
	Negative control†	0	0	0
S suis	Supernatant	4574	4110	3654
NT1097925§	Biofilm	377	258	256
	Positive control†	5035	3713	3031
	Negative control†	0	0	0
	= -			

^{*} For each organism, the virus suspension was added to a bacterial culture and incubated in a standard microtiter plate assay for biofilm formation. Testing for PRRSV was conducted daily, using a commercial PCR kit (EZ-PRRSV MPX 4.0; Tetracore, Rockville, Maryland), for up to 3 days in the supernatant (liquid phase above the biofilm) and in the biofilm attached to the plastic surface.

- † Positive control, no bacteria, virus only; negative control, no virus, bacteria only.
- † This strain produces a biofilm in the presence of fibrinogen.8
- § This strain does not require fibrinogen to produce a biofilm.⁹

Results of experiments using the CellTiter-Blue cell viability assay to test the antibacterial efficacy of both disinfectants against *A pleuropneumoniae* biofilms clearly indicated that both disinfectants were effective to decrease the metabolic activity of *A pleuropneumoniae* under the study conditions. Overall, metabolic activity was lower by 90% to 100% in the biofilm samples treated with disinfectants than in the non-treated samples (data not shown), suggesting that large numbers of bacterial cells had died. It is important to note that the disinfectants killed the bacterial cells but did not remove the biofilms.

Discussion

The effects of biofilms of enteric (E coli or Salmonella) or respiratory (A pleuropneumoniae or S suis) bacterial pathogens on PRRSV and PCV2, two of the most important viruses in the swine industry, were studied. Several control experiments were conducted prior to initiating this pilot project to evaluate, for example, methods for virus recovery from a bacterial biofilm, or to ensure that no traces of disinfectant were left that would affect the cell lines used for viral titration. Overall, our results indicate that a small portion of the viral inoculum persisted in the biofilms for the duration of the experiments, as first determined by qPCR. The amount of PRRSV was too small to attempt

infectious virus quantification by titration on MARC-145 cell lines. However, the amount of PCV2 was greater, and viral titration on NPTr cells was performed, confirming that a proportion of the PCV2 inoculum persisted in the biofilms and remained infectious. A recent publication ¹³ indicated that binding of an enteric virus (poliovirus) to bacterial polysaccharides stabilizes the virions and may offer a selective advantage by enhancing environmental stability.

Although the amounts of virus persisting in bacterial biofilms were relatively low for one of the tested viruses (namely, PRRSV), there was a possibility that once incorporated in a biofilm, these viruses would be protected from disinfectants. Our preliminary results

PCR = polymerase chain reaction; PRRSV = porcine reproductive and respiratory syndrome virus; $TCID_{50}$ = median tissue culture infectious dose.

Table 2: Results of testing for PCV2b in *Actinobacillus pleuropneumoniae*, *Escherichia coli*, *Salmonella*, and *Streptococcus suis* biofilms by virus-specific PCR (expressed as $TCID_{50}/mL$)*

		TCID ₅₀ of PCV2b/mL
A pleuropneumoniae	Supernatant	7.21 × 10 ⁵
	Biofilm	5.63 × 10 ⁴
	Positive control†	9.77×10^5
	Negative control†	0
E coli	Supernatant	4.99×10^5
	Biofilm	5.89×10^4
	Positive control†	9.68×10^5
	Negative control†	0
Salmonella	Supernatant	1.78 × 10 ⁴
	Biofilm	1.74 × 10 ⁴
	Positive control†	7.90 × 10 ⁵
	Negative control†	0
S suis	Supernatant	7.71×10^4
735‡	Biofilm	3.57×10^4
	Positive control†	3.48×10^5
	Negative control†	0
S suis	Supernatant	3.41×10^5
NT1097925§	Biofilm	5.15 × 10 ⁴
	Positive control†	3.48×10^5
	Negative control†	0

- * The virus suspension was added to a bacterial culture and incubated in a standard microtiter plate assay for biofilm formation for 24 hours for *A pleuropneumoniae*, *E coli*, and *S suis*, and for 48 hours for *Salmonella* in order to achieve optimal biofilm formation. The presence of the virus was determined for up to 2 days in the supernatant (liquid phase above the biofilm) and in the biofilm attached to the plastic surface.
- † Positive control: no bacteria, virus only; negative control, no virus, bacteria only.
- † This strain produces a biofilm in the presence of fibrinogen.⁸
- § This strain does not require fibrinogen to produce a biofilm.⁹

PCV2b = porcine circovirus type 2b; PCR = polymerase chain reaction; $TCID_{50}$ = median tissue culture infectious dose.

indicate that bacterial biofilms seem to only slightly reduce the efficacy of disinfectants, which nevertheless remain effective against the virus tested.

To the best of our knowledge, this is the first description of the persistence of two important swine viral pathogens, PRRSV and PCV2b, within bacterial biofilms. This pilot project generated preliminary data important for the swine industry in a new area that certainly deserves to be investigated in more detail. It would be relevant to perform similar experiments with other important swine viruses such as the porcine epidemic diarrhea virus or the porcine deltacoronavirus.

Implications

- PRRSV and PCV2 can associate with bacterial biofilms that are known to be present inside the infected host or in the farm's environment.
- PRRSV and PCV2 can persist within Gram-positive and Gram-negative bacterial biofilms for several days.
- Under the conditions of this study, the efficacy of acid peroxygen and quaternary ammonium-glutaraldehyde commercial disinfectants against PCV2 may be only slightly reduced by the presence of a bacterial biofilm.

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Conflict of interest

None reported.

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Table 3: Detection of PCV2b infectious viral particles (TCID₅₀/mL) in an Actinobacillus pleuropneumoniae biofilm*

TCID₅₀ of PCV2b/mL (log difference from control)

	Control	Quaternary	Acid peroxygen
		ammonium-glutaraldehyde	
Wells without biofilm	10 ^{4.00} †	10 ^{2.90} † (1.10)	10 ^{2.80} † (1.20)
Wells with A pleuropneumoniae biofilm	10 ^{2.60} ‡	10 ^{1.75} ‡ (0.85)	10 ^{1.90} ‡ (0.70)

- * The virus suspension (PCV2b virus strain FMV06-0732) was added to the bacterial culture and incubated in a standard microtiter plate assay for biofilm formation. Porcine circovirus type 2 virions, in the presence or absence of biofilms, were then exposed to acid peroxygen and quaternary ammonium-glutaraldehyde disinfectants at 1% each and for 30 minutes of exposure. Control wells contained no disinfectant. Infectious viral titers were determined using the NPTr cell line. The numbers of infectious viruses in the biofilm attached to the plastic surface were arithmetically greater than the numbers in wells without a biofilm.
- † Number of infectious viruses present in the liquid suspension.
- † Number of infectious viruses present in the biofilm phase only.

PCV2b = porcine circovirus type 2b; $TCID_{50}$ = median tissue culture infectious dose.

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BRIEF COMMUNICATION

Postpartum injection of human chorionic gonadotrophin: Effects on sow ovarian follicles

Jessica Zemitis; Emmy G. Bouwman; P. Langendijk, PhD; W. H. E. J. van Wettere, PhD; R. N. Kirkwood, DVM, PhD, Diplomate ECAR

Summary

Sows received 1000 IU human chorionic gonadotrophin at 24 or 48 hours after farrowing or served as controls. Ovaries were examined ultrasonically at 0, 24, 48, 72, 96, and 120 hours. No sows ovulated by 120 hours, although corpora lutea at 10 days indicated a later ovulation in some sows.

Keywords: sow, farrowing, human chorionic gonadotrophin, ovulation, ultrasound

Received: June 3, 2014 Accepted: November 18, 2014 Resumen - Inyección postparto de gonadotropina coriónica humana: efectos en los folículos ováricos de hembras

Las hembras recibieron 1000 UI de gonadotropina coriónica humana 24 o 48 horas después del parto o sirvieron como controles. Se examinaron los ovarios por ultrasonido a las 0, 24, 48, 72, 96, y 120 horas. Ninguna hembra ovuló a las 120 horas, sin embargo el cuerpo lúteo a los 10 días indicó una ovulación tardía en algunas hembras.

Résumé - Injection post partum de gonadotrophine chorionique humaine: effets sur les follicules ovariens de truies

Des truies reçurent 1000 UI de gonadotrophine chorionique humaine 24 ou 48 heures suivant la parturition ou servirent de témoins. Les ovaires furent examinés par échographie à 0, 24, 48, 72, 96, et 120 heures. Aucune truie n'avait ovulé 120 heures suivant le traitement, bien que les corps jaunes à 10 jours indiquent une ovulation tardive chez quelques truies.

active luteinizing hormone (LH) pulsatility has been observed for up to 78 hours post farrowing, after which suckling-induced inhibition of LH pulsatility takes effect. Further, postpartum sow ovaries have potentially estrogenic medium follicles (4 to 5 mm) and some sows exhibit estrous behavior. However, the postpartum estrus observed at 2 to 4 days post farrowing is anovulatory, likely due to an inability to generate a preovulatory LH surge.

If litters are weaned immediately after farrowing (zero-weaning) so that the suckling-induced inhibitions are removed, estrogenic follicles may continue development, which may trigger estrus and ovulation. ^{1,5,6} However, zero-weaning is also associated with development of cystic follicles and poor subsequent reproductive performance, ^{5,7} a further effect of the inability to mount a preovulatory LH surge. Interestingly, previous workers have provided an exogenous postpartum ovulatory signal in attempts to induce ovulation, as the ovary is still receptive

to exogenous gonadotrophins. Specifically, injection of 1000 IU human chorionic gonadotrophin (hCG) within 24 hours of farrowing induced ovulation in 75% and 41% of sows, respectively. 8,9 In these earlier studies, determination of ovulation was based on detection of serum progesterone concentrations of \geq 5 ng per mL at 7 to 10 days after injection. Although the reasons for the different responses are unknown, an influence of timing of injection cannot be discounted. Similarly, to our knowledge, direct serial observations of ovarian follicular dynamics in individual postpartum sows have not been documented.

If inducing ovulation early in lactation initiates a normal estrous cycle followed by a secondary ovulation, it could result in a novel estrus synchronization protocol. If predictable ovulation can be achieved, it would have great utility as an inexpensive method of postpartum estrus suppression. Such activity would be invaluable under conditions of forced zero-weaning, eg, as a consequence of a disease such as herd

infection with porcine epidemic diarrhea virus. It could also be employed to capture the potential for mating during lactation. The objective of the current study was to determine ovarian follicular dynamics in the immediate postpartum period and the relationship between ovarian follicular status and the response to hCG injection at either approximately 24 or approximately 48 hours post partum.

Material and methods

This study was approved by the University of Adelaide Animal Ethics Committee.

A total of 48 mixed-parity sows (mean \pm standard error [SE], 2.5 \pm 0.2) were selected for convenience and were housed in farrowing crates from 110 days of gestation until weaning. After farrowing, litter sizes were standardized to 10 or 11 (mean \pm SE, 10.9 \pm 0.2 piglets), and piglets were weaned at 28 days post farrowing. During lactation, sows were fed to appetite with a diet formulated to provide 14.3 MJ digestible energy per kg, 12.5% crude protein, and 0.9% total lysine.

Sows were each assigned to one of three treatments by parity. Treatments were intramuscular injection of 1000 IU hCG (Chorulon; MSD Animal Health, Bendigo, Australia) either 24 to 30 hours after farrowing (hCG24; n=16), or 48 to 54 hours after farrowing (hCG48; n=18), or no injection and serving as controls (Control; n=14). Sows farrowing overnight were treated at

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This article is available online at http://www.aasv.org/shap.html.

Zemitis J, Bouwman EG, Langendijk P, et al. Postpartum injection of human chorionic gonadotrophin: Effects on sow ovarian follicles. *J Swine Health Prod.* 2015;23(3):137–139.

9:00 AM the day after farrowing (hCG24) or 2 days after farrowing (hCG48). Sows farrowing during the day (8:00 AM to 4:00 PM) were treated at 24 hours or 48 hours after the end of farrowing.

Transrectal real-time ultrasound (MyLabOne; Esaote Pie Medical, Maastricht, The Netherlands) with an 8-MHz transducer was used to examine ovarian follicle size and number. The ovaries of all sows were scanned at 0, 24, 48, 72, and 96 hours after farrowing, with the hCG48 sows also scanned at 120 hours, to monitor follicle development and determine ovulation. Sows were deemed to have ovulated when pre-ovulatory follicles observed on the previous scan had disappeared. The hCG24 sows were expected to ovulate between 48 and 96 hours after injection, and the hCG48 sows were expected to ovulate between 72 and 120 hours after injection. All sows were also scanned at 10 days of lactation to determine presence of corpora lutea (CLs). Sows were scanned between 7:00 AM and 11:00 AM. For each scan, one ovary was located and scanned from end to end. A video clip of the ultrasound was saved and analyzed for size and number of follicles and presence of CLs.

The data were analysed using SAS (SAS Institute Inc, Cary, North Carolina). In addition to follicle disappearance, evidence of ovulation included the presence of CLs at 10 days. Sows were retrospectively categorised as ovulating or non-ovulating, and the difference between treatments was tested using chi-square. Maximum follicle size was the diameter of the largest follicles at each time point. A generalized linear model was used to compare treatments, and those that ovulated versus non-ovulated in their maximum follicle size, using the following model: $y = \mu + A + day + A^*day + e$, where A is either treatment or ovulation status, μ is estimated overall mean, and e is unexplained error. To determine follicular dynamics, follicles were assigned into two classes: < 5 mm (small) and ≥ 5 mm (large), and number of follicles in each class was counted for each scan. Differences between treatments were considered significant when P < .05.

Results

There were no significant differences between control, hCG24, or hCG48 for parity (parities 3.1 ± 0.4 , 2.4 ± 0.3 , and 2.1 ± 0.2 , respectively) or litter size suckled (10.7 ±0.2 , 10.8 ±0.3 , and 10.9 ±0.2 piglets, respectively). On the basis of presence of

CLs, none of the control sows ovulated, while five of the 16 hCG24 sows and four of the 18 hCG48 sows had CLs, indicating ovulation had occurred. However, ultrasound examinations showed that ovulation was not detected by 120 hours post farrowing, indicating that ovulation occurred later and was not directly induced by the hCG injection.

At the first ovarian scan immediately following farrowing, 21% of the sows had one or more follicles ≥ 5 mm. At 24, 48, 72, and 96 hours, the percentages of sows with follicles ≥ 5 mm were 38%, 32%, 29%, and 39%, respectively. The diameters of the largest follicles were between 4.9 and 9.0 mm at the first scan post farrowing. Sows in the Control group exhibited follicle growth of 0.68 mm during the 24 hours after farrowing, but then follicle size decreased by 0.4 mm between 24 and 96 hours post farrowing (Figure 1). In contrast, hCG24 sows experienced follicle growth of 1.0 mm from 48 to 96 hours, while hCG48 sows exhibited follicle growth of 1.2 mm from 72 hours to 120 hours. At 72 and 96 hours, follicle diameter was larger (P < .05) in sows destined to ovulate than in sows that did not ovulate.

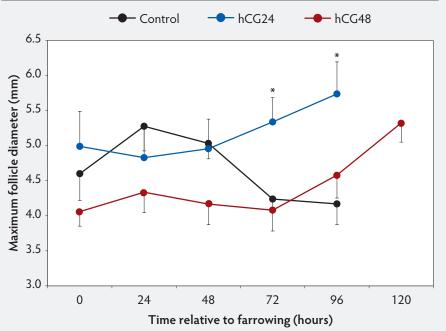
Discussion

Injecting sows with 1000 IU hCG in the immediate postpartum period resulted in eventual ovulation in 33% and 22% of

hCG24 and hCG48 sows, respectively, as indicated by presence of CLs at 10 days. No control sows had evidence of ovulation. The current data differs from previous reports of 71%8 and 41%9 of sows ovulating in response to an injection of 1000 IU hCG within 24 hours of farrowing. Compared to the results of previous studies, hCG treatment in the present study was considerably less successful. Previous studies used only progesterone concentrations at 7 to 10 days after farrowing to determine whether ovulation had taken place or not. For the current study, ultrasound was employed to determine occurrence of ovulation on the basis of follicle disappearance and observation of luteal structures at 10 days post partum. Our failure to detect ovulation in the immediate postpartum period likely means that hCG is not capable of inducing ovulation at this time and that previous reports indicating ovulation based on blood progesterone concentrations were incorrect. The LH pulsatility immediately postpartum is very active, and it is possible that soon after farrowing, the follicular LH receptors may be downregulated. This would preclude an ovulation in response to an exogenously supplied ovulatory LH surge.

The follicular dynamics in the control sows were similar to what has been found in

Figure 1: Diameter of the largest follicles as determined by transrectal ultrasonography during the 120 hours after farrowing in sows receiving 1000 IU human chorionic gonadotropin (hCG) at approximately 24 hours (hCG 24; n = 16) or 48 hours (hCG48; n = 18) after farrowing, or not treated (Controls; n = 14). Asterisks (*) indicate data points where maximum follicle size differs between hCG24 sows and Control sows (P < .05; generalized linear model).



previous studies; those studies found that follicles were no bigger than 5 mm in early lactation, ¹ while others found that follicles did not exceed 3 to 4 mm in diameter. ¹⁰⁻¹² An average follicle diameter of 4.6 mm after farrowing has also been documented, which declined to 2.6 mm over a week. ¹³ These data are consistent with the results seen in our control sows, which showed a decrease in follicle size over the 5 days following farrowing.

In cyclic gilts and sows, follicles of approximately 3 to 4 mm in size grow primarily in response to LH activity. 14 Of the sows that had evidence of ovulation in the present study, all had follicles approximately 4 mm in diameter at the time of injection. We did observe follicle growth in hCG-treated sows, in contrast to controls. Therefore, the CLs observed at 10 days may be a result of hCGinduced follicle growth with subsequent spontaneous ovulation prior to 10 days. Of the 34 sows that received an injection, 23 were recorded as having an increase in follicle size, follicle numbers, or both. However, other sows had similar follicle size and number at the time of injection, but were unable to achieve ovulation. The factor(s) that determined why some ovulated but others did not remain unknown.

Implication

Under the conditions of this study, an immediate ovulatory response of farrowed sows to hCG cannot be confirmed, and therefore this treatment modality will not predictably control postpartum ovarian function.

Acknowledgements

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Conflict of interest

None reported.

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Brief communication

Susceptibility of 45 recent French field isolates of Streptoccocus suis to florfenicol

Guillaume Lequeux, DVM, MSc; Eric Le Drean, DVM, PhD

Summary

To investigate occurrence of acquired resistance, minimum inhibitory concentrations of florfenicol for *Streptococcus suis* isolated in France between 2011 and 2014 were determined. No acquired resistance to florefenicol was observed among recent field isolates of *S suis* after more than 10 years of use of this antibiotic.

Keywords: swine, swine respiratory disease, *Streptococcus suis*, florfenicol

Received: July 7, 2014 Accepted: October 28, 2014 Resumen - Susceptibilidad al florfenicol de 45 aislamientos franceses de campo recientes de *Streptoccocus suis*

Para investigar la ocurrencia de la resistencia adquirida, las concentraciones mínimas inhibitorias de florfenicol para aislamientos de *Streptoccocus suis* en Francia entre 2011 y 2014 fueron determinados. No se observó resistencia adquirida al florfenicol entre aislamientos de campo recientes de *S suis* después de más de 10 años de uso de este antibiótico.

Résumé - Sensibilité au florfénicol de 45 isolats récents de *Streptococcus suis* provenant de France

Afin d'étudier l'occurrence de résistance acquise envers le florfénicol, on détermina les concentrations minimales inhibitrices d'isolats de *Streptococcus suis* obtenus entre 2011 et 2014 d'élevages en France. Aucune résistance acquise au florfénicol ne fut observée parmi les isolats récents de *S suis* après plus de 10 ans d'utilisation de cet antibiotique.

Streptococcus suis is a major pathogen in swine production, causing meningitis, arthritis, septicemia, bronchopneumonia, polyserositis, and endocarditis. ¹ It is also recognized as an important zoonotic agent. ²

A florfenicol concentrate solution is labelled in the United States for treatment of swine respiratory disease (SRD) associated with several bacterial pathogens, including S suis. Treatment is administered by the oral route through drinking water. In France, florfenicol is approved for treatment and control of respiratory disease caused by the major pathogens Actinobacillus pleuropneumoniae and Pasteurella multocida, but S suis is not included in the claim. However, S suis may occur simultaneously or sequentially with such bacteria in SRD and is also found in the upper respiratory tracts of healthy animals.³ Thus S suis may be exposed to florfenicol during treatment of animals suffering from SRD caused by bacterial pathogens.

The objective of this study was to determine susceptibility to florfenicol of recent *S suis*

field isolates from pig herds in western France to determine whether acquired resistance has emerged. Minimum inhibitory concentrations (MICs) were determined to provide epidemiological survey data.

Materials and methods

Bacterial isolates

Bacteria were isolated at the Institut en Santé Agro-Environnement laboratory (Public Veterinary Diagnostic Laboratory, Fougeres, France) between 2011 and 2014 from samples submitted for disease diagnosis in piglets from herds located in the west of France (mainly Ille-et-Vilaine, but also in Brittany, Pays de la Loire, and Normandy). Isolation, identification, and serotyping of isolates were conducted by conventional bacteriological methods: culture, Gram staining, biochemical tests (Api 20 Strep strip; BioMérieux, Marcy l'Etoile, France) and serotyping (*S suis* antisera; LDA 22, Ploufragan, France).

All isolates were stored at < -60°C in brainheart broth with 15% glycerol until MIC determination. Determination of MICs was performed on independent *S suis* isolates from an epidemiological point of view (ie, no more than one isolate per year per herd).

MIC determination

Minimal inhibitory concentrations were determined using a broth microdilution method. Testing was performed according to Clinical and Laboratory Standards Institute guidelines (CLSI VET01-A4⁴ and VET01-S2⁵). Briefly, after isolates were incubated overnight on agar plates and purity of the cultures was confirmed, the direct colony suspension method was used. Bacterial suspensions were adjusted in cation-adjusted Mueller-Hinton broth supplemented with 2.5% lysed horse blood. The final concentration in the 50 µL of bacterial suspension added per microtiter plate well was approximately 5×10^5 colony forming units (CFU) per mL. Minimum inhibitory concentrations were determined using 96-well microtiter plates containing dehydrated antibiotic (CMP1ASPV, florfenicol custom veterinary susceptibility plate format; Trek Diagnostic Systems Inc, Cleveland, Ohio). After incubation for 24 hours at 35°C (standard deviation 2°C) in ambient air, the MIC was recorded as the lowest concentration of florfenicol that completely

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Lequeux G, Le Drean E. Susceptibility of 45 recent French field isolates of *Streptoccocus suis* to florfenicol. *J Swine Health Prod.* 2015;23(3):140–142.

inhibited growth of the organism in wells, as detected by the unaided eye.

Quality controls

As directed in CLSI guidelines, 4,5 Enterococcus faecalis ATCC 29212 and Streptococcus pneumoniae ATCC 49619 were used as reference strains for MIC quality controls each day of testing, with acceptable quality control ranges 2 to 8 μ g per mL and 1 to 4 μ g per mL, respectively. A colony count of the inoculum for each plate was performed to ensure that the final inoculum in wells approximated 5 \times 10⁵ CFU per mL.

Results

The 45 *S suis* isolates used in the study were from routine submissions to the Public Veterinary Diagnostic Laboratory, from piglets 4 to 9 weeks old suffering from respiratory disease (19 isolates; 42%), endocarditis (9 isolates; 20%), septicemia (8 isolates; 18%), meningitis (5 isolates; 11%), and arthritis (4 isolates; 9%). Isolates belonged to different serotypes: type 2 (38% of isolates), non-typeable (24%), 7 (13%), 1,2 (7%), 3 (7%), 1 (4%), 8 (4%), and 9 (2%). Minimum inhibitory concentrations of florfenicol ranged from 0.5 to 2 μ g per mL; MIC₅₀ and MIC₉₀ were 1 and 2 μ g per mL, respectively, confirming the previous data, as shown in Table 1.

Discussion

All isolates included in this study were considered to be susceptible to florfenicol,

according to CLSI VET01-S2-approved breakpoints for *S suis*, which are $\leq 2 \mu g$ per mL (susceptible), $4 \mu g$ per mL (intermediate), and $\geq 8 \mu g$ per mL (resistant). Data provided by this study are consistent with results of previous studies in which resistance was not detected among isolates collected in France until 2002¹² and in Germany between 2000 and 2005. 8-10

Streptococcus suis isolates classified as intermediate to florfenicol have seldom been isolated in Europe¹¹ or North America.⁷ According to the studies of Callens et al⁶ and Portis et al,⁷ resistance to florfenicol is seldom if ever reported (one isolate among 331 European and approximatively 2000 American isolates tested), whereas the susceptibilities of S suis isolates to several antibiotics (erythromycin, lincomycin, penicillin, tiamulin, tetracycline, tilmicosin, and tylosin) have dramatically decreased in Europe over the past few years.⁶

Distribution of isolates among serotypes is in accordance with previous French data, but with a higher percentage of non-typeable isolates and a lower percentage of serotype 9.¹³ Wisselink et al¹¹ could not show an association between the serotype of an isolate and its susceptibility pattern. The number of isolates in this study was too small to confirm this result.

Although no resistance to florfenicol was found among the *S suis* isolates tested in this study, the authors do not recommend extralabel use of florfenicol.

Implication

Under the conditions of this study, *S suis* isolates collected from piglets in the west of France between 2011 and 2014 were not resistant to florfenicol.

Conflict of interest

None reported.

Disclaimer

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Table 1: Florfenicol MIC₅₀, MIC₉₀, and MIC ranges reported for Streptococcus suis isolates*

		Concentrations			
	S suis isolation	MIC ₅₀ (μg/mL)	MIC ₉₀ (μg/mL)	MIC ranges (μg/mL)	of florfenicol (μg/mL) [‡]
Present study	2011-2014	1	2	0.5-2	0.125-128
Callens et al ⁶	2010	ND	ND	0.5-8	0.03-128
Portis et al ⁷	2007-2010	2	2	0.06->32	0.06-32
	2001-2006	1	2	0.06->32	0.06-32
Schwarz and Kehrenberg ⁸	2000-2005	1	2	0.25-2	0.125-128
Kehrenberg et al ⁹	2002-2003	1	2	0.25-2	0.125-128
Priebe and Schwarz ¹⁰	2000-2001	1	2	0.25-2	0.125-128
Wisselink et al ¹¹	1987-1997	ND	ND	0.5-4	0.06-32

^{*} Areas of isolation: West of France (present study); Belgium (Callens); North America: United States and Canada (Portis); Germany (Schwarz, Kehrenberg, Priebe); Belgium, UK, France, Italy, Spain, Germany, and The Netherlands (Wisselink).

[†] MIC determination methods: Broth microdilution for the present study and all references except Callens⁶ (agar dilution).

[‡] Range of concentrations of florfenicol tested.

MIC = Minimum inhibitory concentration; ND = not determined

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- * Non-refereed references.



CONVERSION TABLES

Weights and measures conversions

	weights and measures conversions						
Common (US)	Metric	To convert	Multiply by				
1 oz	28.35 g	oz to g	28.4				
1 lb (16 oz)	453.59 g	lb to kg	0.45				
2.2 lb	1 kg	kg to lb	2.2				
1 in	2.54 cm	in to cm	2.54				
0.39 in	1 cm	cm to in	0.39				
1 ft (12 in)	0.31 m	ft to m	0.3				
3.28 ft	1 m	m to ft	3.28				
1 mi	1.6 km	mi to km	1.6				
0.62 mi	1 km	km to mi	0.62				
1 in ²	6.45 cm ²	in^2 to cm^2	6.45				
0.16 in ²	1 cm ²	cm^2 to in^2	0.16				
1 ft ²	0.09 m^2	$\mathrm{ft}^2\mathrm{to}\mathrm{m}^2$	0.09				
10.76 ft ²	1 m ²	m^2 to ft^2	10.8				
1 ft ³	0.03 m^3	$\mathrm{ft}^3\mathrm{to}\mathrm{m}^3$	0.03				
35.3 ft ³	1 m ³	m^3 to ft^3	35				
1 gal (128 fl oz)	3.8 L	gal to L	3.8				
0.264 gal	1 L	L to gal	0.26				
1 qt (32 fl oz)	946.36 mL	qt to L	0.95				
33.815 fl oz	1 L	L to qt	1.1				

Tem	perature	equiva	lents (ap	prox)	

remperature equivalents (approx)						
°F	°C					
32	0					
50	10					
60	15.5					
61	16					
65	18.3					
70	21.1					
75	23.8					
80	26.6					
82	28					
85	29.4					
90	32.2					
102	38.8					
103	39.4					
104	40.0					
105	40.5					
106	41.1					
212	100					
°F = (°C × 9/5) + 32						

 $^{^{\}circ}F = (^{\circ}C \times 9/5) + 32$ $^{\circ}C = (^{\circ}F - 32) \times 5/9$

Conversion	chart	lh to	kσ	(approx)	١
COLIVEISION	Ciiai C	וט נט	KK I	(αρριολ	,

	ii ciiai q ib to i	(approx)
Pig size	Lb	Kg
Birth	3.3-4.4	1.5-2.0
Weaning	7.7	3.5
	11	5
	22	10
Nursery	33	15
	44	20
	55	25
	66	30
Grower	99	45
	110	50
	132	60
Finisher	198	90
	220	100
	231	105
	242	110
	253	115
Sow	300	135
	661	300
Boar	794	360
	800	363
1. 10/	20.1	

 $^{1 \}text{ tonne} = 1000 \text{ kg}$

 $^{1 \}text{ ppm} = 0.0001\% = 1 \text{ mg/kg} = 1 \text{ g/tonne}$

¹ ppm = 1 mg/L

Thiamine-responsive neurological disorder of swine

Sara D. Hough, DVM; Samuel H. Jennings, DVM, Diplomate ACVP; Glen W. Almond, DVM, PhD

Summary

This report describes a thiamine-responsive neurological disease and the methodology leading to its diagnosis. The initial case involved one nursery farm. Approximately 5% of pigs at 5 to 7 days after weaning exhibited central nervous system signs. Over the next 3 weeks, 16 of the company's 41 nursery farms had pigs with similar clinical signs. One month later, neurologic signs were observed in unweaned piglets in several sow farms. Pigs were weaned at approximately 19 days and moved to off-site nurseries. Live

pigs and fresh and formalin-fixed samples from acutely affected pigs were sent to diagnostic laboratories. Feed samples were submitted for mycotoxin and nutrient analyses. Initial reports revealed no precise cause of the neurological condition; however, polioencephalomalacia (PEM) subsequently was identified in affected pigs. A field trial determined the response to treatment with atropine, a vitamin A, D, and E preparation, or vitamin B12 plus thiamine. Pigs treated with thiamine recovered from the neurological condition. Upon implementation

of thiamine injections on a company-wide basis, neurological signs associated with PEM were no longer evident. The authors do not recommend routine thiamine injections under normal circumstances. In this case, compromised dietary thiamine levels during feed manufacturing possibly contributed to the PEM.

Keywords: swine, polioencephalomalacia, thiamine, neurological signs

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Resumen - Desorden neurológico porcino con respuesta a la tiamina

Este reporte describe la enfermedad neurológica con respuesta a la tiamina y la metodología que llevó a su diagnóstico. El caso inicial involucró un sistema de destete. Aproximadamente 5% de los cerdos de 5 a 7 semanas después del destete exhibieron signos relacionados con el sistema nervioso central. Durante las siguientes 3 semanas, 16 de las 41 granjas de destete de la compañía tuvieron cerdos con signos clínicos similares. Un mes después, se observaron signos neurológicos en lechones no destetados en varias granjas de pie de cría. Los cerdos se destetan a los 19 días aproximadamente y son trasladados a destetes fuera de sitio. Se enviaron al laboratorio de diagnóstico cerdos vivos y muestras frescas y fijadas en formalina de cerdos afectados de forma aguda. Se enviaron muestras de alimento para análisis de micotoxinas y nutrientes. Los reportes iniciales no revelaron una causa precisa de la enfermedad neurológica;

sin embargo, subsecuentemente se identificó polioencefalomalacia (PEM por sus siglas en inglés) en los cerdos afectados. Una prueba de campo determinó la respuesta al tratamiento con atropina, una preparación de vitamina A, D, y E, o vitamina B12 más tiamina. Los cerdos tratados con tiamina se recuperaron de la enfermedad neurológica. Después de la implementación de la inyección de tiamina en todos los cerdos de la compañía, los signos neurológicos asociados con el PEM ya no se manifestaron. Bajo circunstancias normales, los autores no recomiendan las inyecciones rutinarias de tiamina. En este caso, los niveles de tiamina en la dieta, afectadas durante la preparación del alimento, posiblemente contribuyeron al PEM.

Résumé - Maladie neurologique des porcs répondant à la thiamine

Le présent rapport décrit une maladie neurologique répondant à la thiamine et la

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méthodologie menant à son diagnostic. Le cas initial impliquait une pouponnière. Cinq à 7 jours suivant le sevrage environ 5% des porcs démontraient des signes d'atteinte du système nerveux central. Durant les 3 semaines qui suivirent, dans 16 des 41 pouponnières que possèdent l'entreprise des porcs ont présenté des signes cliniques similaires. Un mois plus tard, des signes neurologiques furent observés chez des porcelets non-sevrés dans plusieurs des fermes de maternité. Les porcs furent sevrés à environ 19 jours et déménagés à une pouponnière hors-site. Des porcs vivants et des échantillons de tissus frais et fixés dans la formaline provenant de porcs affectés de manière aiguë furent acheminés à des laboratoires de diagnostic. Des échantillons de nourriture furent soumis pour analyse des nutriments et détection de mycotoxine. Les rapports initiaux ne démontraient aucune cause précise de la condition neurologique; toutefois, une polioencéphalomalacie (PEM) subséquente fut identifiée chez les porcs atteints. Un essai clinique a déterminé la réponse à un traitement avec de l'atropine, une préparation de vitamines A, D, et E, ou de la vitamine B12 plus thiamine. Les porcs traités avec de la thiamine ont récupéré de la condition neurologique. Avec la mise-enplace d'injections de thiamine à l'ensemble des animaux de la compagnie, les signes neurologiques associés à la PEM n'étaient plus évidents. Les auteurs ne recommandent pas l'injection de thiamine sur une base routinière dans des circonstances normales. Dans le cas présent, des niveaux de thiamine alimentaire déficients durant la préparation de l'aliment ont possiblement contribué à la PEM.

Pem olioencephalomalacia (PEM) is recognized as an important neurologic disease of ruminants. In contrast to PEM in ruminants, thiamine (vitamin B1) deficiencies, and specifically neurologic signs associated with thiamine deficiencies, are rarely reported for swine. Anorexia, reduced weight gain, occasional vomiting, and sudden death are considered the common clinical signs associated with thiamine deficiency in pigs. In addition, thiamine supplementation of modern pig diets provides limited benefit, as primary feed ingredients contain sufficient thiamine to meet the requirements of pigs. 3.4

Thiamine depletion requires considerable time in pigs, and it may take up to 35 days for pigs to exhibit non-neurologic clinical signs. ^{5,6} Dietary deficiency of thiamine apparently is rare, with a few notable case reports; ^{7,8} however, excessive sulfur intake as a result of diet manipulation was noted as a potential cause of thiamine deficiency. ⁶ The precise number of cases of thiamine deficiency in the modern pork industry is unknown.

When it comes to the clinical presentation of disease outbreaks, nutritional deficiencies and alterations to the diet are often low on the list of differential diagnoses. This report describes a thiamine-responsive neurological disease and the diagnostic methodology that led to its discovery.

Clinical description

The chronological events are important to comprehend the changes and extent of this particular case. Thus, the clinical description reviews the events as the clinical signs evolved in several different farms in a production system with over 100,000 sows. All farms were PQA Plus certified, following the guidelines provided by the National Pork Board, prior to the clinical problems.

In February 2012, approximately 5% of the pigs in one of the company's nursery farms exhibited central nervous system signs, with onset of clinical signs 5 to 7 days after weaning at approximately 19 days of age. Clinical signs included ataxia and standing with front legs splayed and head extended, stargazing, trembling, and hyperesthesia (Figure 1). Some pigs entered into lateral recumbency and were unable to rise. Pigs were identified as affected by stimulating them to move about the pens. The affected pigs ran into feeders, cup drinkers, penning

Figure 1: Neurologic signs, such as ataxia, splayed front legs, stargazing, trembling and hyperesthesia, were observed in pigs 5 to 7 days after weaning in one nursery farm. Within 3 weeks, similar neurologic signs were noted in pigs at several nursery farms. This figure illustrates the neurologic posture in a nursery pig exhibiting typical clinical signs of the case.



material, and other pigs, suggesting another undefined problem. These pigs proceeded to exhibit other previously described neurologic signs for PEM. Mortality of affected pigs was 100%. No consistent gross lesions were identified on necropsy of five pigs.

Concurrent to the clinical signs in the nursery, a separate flow of feeder pigs was placed into a finisher barn. The feeder pigs were from a different nursery and were company owned. Their feed was processed in the same feed mill as the feed for the nursery pigs. At placement, 43 animals (approximately 10%) had died on the truck during transport and an additional 25 pigs (approximately 6%) became dyspneic, vocalized, and began to die within 2 hours of being placed into pens. The pens were not crowded and the ambient temperature was 11°C. Necropsy of 20 pigs revealed severe pulmonary edema and copious amounts of serous pleural fluid (Figure 2). Most pigs also had moderate liver congestion, and three pigs had cranioventral pneumonia and mildly edematous mesentery. Samples submitted to the Iowa State Veterinary Diagnostic Laboratory included fresh and formalin-fixed samples of lung, heart, lymph node, liver, spleen, kidney, intestine, brain, and tonsil, as well as serum from affected and unaffected animals.

Over the next 3 weeks, the prevalence of the neurologic signs in nursery pigs increased to involve 16 of the 41 nurseries in the company's North Carolina system. Signs also were observed in weaned pigs from North Carolina that were sent to company-owned,

wean-to-finish facilities in Iowa. One month after the index case, the same clinical signs were seen in a North Carolina sow farm in 23-day-old pigs, 2 days before weaning. Within the subsequent week, the neurologic signs in more sow farms began to be observed in pigs just prior to weaning. The incidence of neonatal diarrhea also increased in numerous sow farms across the system.

Two nurseries that had not experienced the neurologic signs in young pigs began to have a sudden increase in mortality in pigs 2 to 3 weeks after placement. As in the index feeder-pig case, these pigs showed few clinical signs other than dyspnea before dying. Also, some pigs appeared to be vomiting. Necropsy revealed severe pulmonary edema and copious amounts of serous pleural effusions. Injections of isoflupredone acetate, ceftiofur, enrofloxacin, and florfenicol, as well as various water medication strategies (soluble penicillin, chlortetracycline, and tiamulin) did not prevent clinical signs or reduce their severity.

Losses

A marked increase in weekly nursery mortality (Figure 3) occurred during late March and April, representing the loss of approximately 12,000 pigs in the production system during the period, compared to normal mortality levels.

Initial diagnostic tests

A differential list of causes for the neurologic syndrome included *Streptococcus suis*, *Haemophilus parasuis*, water deprivation, edema

disease due to enterotoxigenic *Escherichia coli*, organophosphate toxicosis, nutritional imbalance, and porcine enterovirus and hemagglutinating encephalomyelitis virus. A differential list of causes for fatal pulmonary edema included fumonisin toxicosis, porcine circovirus, influenza, and porcine reproductive and respiratory syndrome (PRRS) virus.

Fresh and formalin-fixed samples from acutely affected pigs were sent to diagnostic laboratories, while samples of the feed were submitted for the isolation, enumeration, and identification of any species of molds detected (JKM Lab, Mount Prospect, Illinois), toxin testing (vomitoxin, aflatoxin, zearalenone, fumonison, 15 trichotheces) at North Dakota State University Veterinary Diagnostic Laboratory (Fargo, North Dakota), organophosphate testing at Diversified Laboratories (Chantilly, Virginia), and heavy-metal screening (arsenic, cadmium, cobalt, chromium, lead, mercury) at Carolina Analytical Services (Bear Creek, North Carolina). Feed samples were also tested at the feed-mill laboratory for fumonisins, deoxynivalenol, and zearalenone. Live pigs displaying clinical signs (n = 29) also were submitted to various diagnostic laboratories.

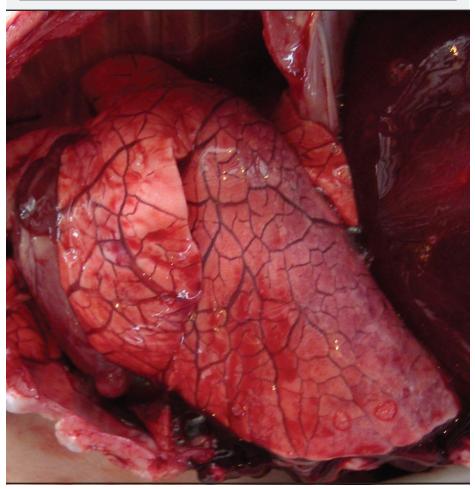
Feed evaluation

All nursery feed was pelleted. On the premise that the neurologic signs were strikingly similar to PEM in cattle and the preliminary findings that injections of thiamine elicited positive responses, feed samples were evaluated for thiamine concentrations. Diagnostic evaluation of the complete feed revealed a thiamine level of 0.62 mg per kg, which represents only 18% of the expected 3.5 mg per kg. Mycotoxin evaluation showed that all feed samples were negative for aflatoxin, fumonisins, and deoxynivalenol. Samples were negative for tremorgens, pesticides, and heavy metals.

Initial postmortem findings

Fourteen nursery-pig submissions (one to five pigs per submission) initially were submitted to diagnostic laboratories. Brain lesions were not identified in the initial pig submissions (Table 1). Subsequent submissions included pigs with meningeal thickening and neutrophilic inflammation of the brain, as well as cortical necrosis and edema suggestive of *S suis* or *H parasuis*. There was growth of *S suis* from a brain swab of an individual pig, but other swabs and tissues did not yield bacterial growth. The ninth

Figure 2: Severe pulmonary edema in a feeder pig. In addition to the neurologic signs observed in pigs at several nursery farms (Figure 1), pulmonary edema was a common finding in several pigs.



nursery-pig submission revealed lesions of PEM of the cerebral cortex, and the lesion was seen in the remaining six nursery pigs submitted. Heavy metal testing was unremarkable in all cases.

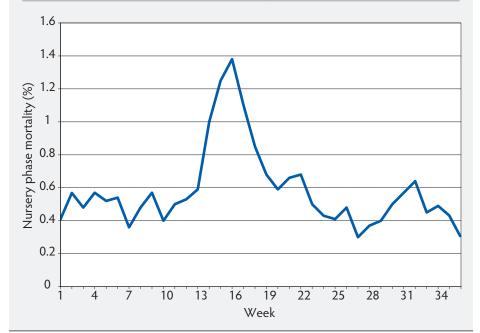
Severe pulmonary edema was observed in one finishing-pig submission. Microscopic lesions confirmed pulmonary edema as well as extensive congestion of cardiac and hepatic tissue. Evaluation of serum yielded hypovitaminosis D and increased alkaline phosphatase. No etiologic agent was suggested. Among three wean-to-finish submissions were two revealing meningeal edema and bronchopneumonia. Pigs were diagnosed with PRRS and hypovitaminosis A and D. Lesions of PEM and neuronal necrosis, as well as interstitial pneumonia, were observed in the third finishing pig submission. Brain sodium levels were tested and were considered low (1439 mg per kg) and below the threshold of 2000 mg per kg for salt toxicosis.

Among three sow-farm submissions, two piglets had focal necrosis of the cerebral cortex and non-suppurative meningitis of unknown etiology. Samples were negative for porcine enterovirus and hemagglutinating encephalomyelitis virus. In the third submission, there were no lesions, and cholinesterase activity in the brain was normal.

Summary of diagnostic findings

The diagnostic findings included 23 accessions (Table 1). Complete necropsy results were obtained from 23 pigs at laboratories other than North Carolina State University (NCSU). Of these pigs, 15 (61%) were identified as having brain lesions: six (26%) were described as having PEM or cortical necrosis of the cerebrum, three (13%) had mononuclear inflammation, three (13%) had neutrophilic or mixed inflammation, and three (13%) had other non-inflammatory brain lesions, including edema, hemorrhage, or neovascularization. One pig with PEM also had areas of mixed meningeal inflammation.

Figure 3: Neurologic signs were observed in pigs 5 to 7 days after weaning in several nursery farms in North Carolina. As a result of the neurologic condition, mortality increased in the nursery farms of the company. This figure depicts weekly nursery mortality for 38 weeks (January through September 2012).



The majority of pigs submitted for diagnostic evaluation were positive for PRRS virus by PCR testing. In addition, some pigs tested positive for swine influenza virus or rotavirus.

Postmortem findings from North Carolina State University

Eight 16- to 18-day old pigs (five females, three males) ranging from 3.8 to 7.0 kg (average 5.8 kg) and showing neurological signs, were examined at NCSU immediately after euthanasia. Six of the eight pigs (75%) exhibited moderate to marked multifocal laminar cerebrocortical necrosis that fluoresced under 365-nm ultraviolet (UV) light from a Wood's lamp. Histologically, the cerebral cortex contained multiple relatively well-demarcated zones of coagulative necrosis, most severe along the grey-white interface of the frontal lobe, but also present in the parietal lobe (Figures 4, 5, and 6). In the early stages or in mild cases, lesions were often limited to middle and deep portions of cerebrocortical sulci, with progression to involve entire gyri. Affected areas had rarefied neuropil with prominent capillaries lined by hypertrophied endothelial cells. These vessels, and vessels of the overlying meninges, were often cuffed by edema and occasionally by one to three layers of lymphocytes, plasma cells, and very rare eosinophils (Figure 7). Neurons in these

areas were frequently necrotic with shrunken angular profiles, cytoplasmic eosinophilia, and loss of Nissl substance (Figures 5 and 7). Alzheimer type II cells were also frequently present individually or in pairs or clusters of up to four cells with enlarged, glassy nuclei and peripheralized chromatin (Figure 6). Vacuolation of the periventricular grey matter of the brainstem was seen in five pigs. Three of these five pigs also had infiltration of Alzheimer type II cells in these areas, with neuronal necrosis at this location in two pigs (Figures 8 and 9).

Treatment trial

While investigators waited for the final diagnostic results from NCSU, 24 pigs exhibiting clinical signs in one facility were placed in three experimental treatment groups (n = 8 pigs per group). Pigs in Group 1 were treated intramuscularly (IM) with 5 mg of atropine sulfate. Pigs in Group 2 received (IM) 1 mL of a product containing vitamin A (50,000 IU), vitamin D (50,000 IU), and vitamin E (500 IU). Pigs in Group 3 were injected IM with 1 mL (100 mg) of vitamin B12 and 1 mL (100 mg) of thiamine (vitamin B1). The following day, each pig was assessed. The pigs injected with atropine were still alive but recumbent. The pigs injected with the fat soluble vitamin mixture were also recumbent. The pigs

injected with the vitamin B compounds were no longer recumbent and appeared to be eating and drinking. Subsets of pigs at two other farms were then injected with thiamine alone and similar positive results were observed. A solution of mixed B vitamins, which included 200 mg thiamine per mL, was distributed to all the nurseries and sow farms for treatment and prophylactic administration to weaning-age pigs and sows. Upon implementation of the thiamine injections, neurological signs associated with the thiamine-induced PEM were no longer evident.

Discussion

Although PEM has been reported in a variety of species, including foxes, mink, dogs, 10,11 cats, 11,12 and camels, 13 few reports historically described thiamine-responsive PEM in pigs. The most commonly described condition for laminar cortical necrosis in the pig is salt toxicosis; however, due to the widespread nature of the epidemic, including several farms, and the paucity of eosinophils histologically, salt toxicosis-water deprivation was considered to be an unlikely cause of the condition. In addition, water availability was not problematic on any of the affected farms.

Polioencephalomalacia is best documented in ruminants. In cattle, PEM is the result of thiamine deficiency (due to thiaminaseproducing bacteria or plants or to true dietary deficiency), sulfur intoxication, lead poisoning, or water deprivation. 14 In ruminants, PEM is characterized by laminar cortical necrosis, with necrotic areas grossly exhibiting yellow-green autofluorescence under 365-nm ultraviolet light. This characteristic was historically attributed to the accumulation of ceroid-lipofuscin in lipophages subsequent to lipid degeneration in injured neuronal cell membranes. However, more recently, it was proposed that intracytoplasmic subunit c of mitochondrial ATP synthetase may be responsible for the autofluorescence. 14 Some cases also exhibit focal symmetric necrosis in the thalamus, colliculi, or brainstem; however, this is more typical of thiamine deficiency in carnivores. 15 Most of the pigs examined at NCSU in this series exhibited both laminar cortical necrosis and degenerative changes in the periventricular grey matter.

The proposed mechanism for sulfur toxi-

Table 1: Diagnostic results obtained from submissions of one or more pigs to diagnostic laboratories*

Date	Farm	Pig age (weeks)	Brain lesions	Other lesions
22 Feb	Index NC case, Nursery 1	4	None	Atrophic enteritis, multifocal dilation of colonic crypts
22 Feb	Index NC case, Finisher 1	10	None	Pulmonary edema, cardiac congestion, hepatic congestion
22 Feb	Nursery 2	4	None	Villous atrophy, ileum
29 Feb	Nursery 3	3	Brain hemorrhage	Epicarditis, interstitial pneumonia
29 Feb	Nursery 4	4	None	Focal myocardial hemorrhage, interstitial pneumonia
7 Mar	Nursery 5	4	Meningeal thickening and neutrophilic inflammation	Villous atrophy of ileum; broncho- and interstitial pneumonia
9 Mar	Nursery 6	3	Purulent meningitis	Interstitial pneumonia, glomerulonephritis
21 Mar	Index NC case, Sow Farm 1	3	Focal neovascularization in cerebral cortex	Lymphocytic gastritis, villous atrophy, jejunum
23 Mar	Sow Farm 2	3	None	Nephritis
23 Mar	Nursery 7	4	Cortical necrosis with edema	Necrotizing bronchiolitis, fibrinous pleuritis; thickened colonic mucosa
27 Mar	Index IA case, Wean-to-finish 1	3	Meningeal edema, perivascular hemorrhage, rare neutrophils	Villus blunting, small intestine
27 Mar	Wean-to-finish 2	3	None	Bronchopneumonia
27 Mar	Nursery 8	4	Focal areas of malacia, gliosis, and necrosis	Atrophic enteritis
27 Mar	Nursery 7, 2 nd set of samples	3	Perivascular lymphocytic cuffing and subdural congestion	None
28 Mar	Nursery 9	4	Polioencephalomalacia, cerebral cortex; leukoencephalomalacia	Villous atrophy, jejunum
28 Mar	Sow Farm 1, 2 nd set of samples	3	Non-suppurative meningitis	None
28 Mar	Nursery 10	4	Multifocal polioencephalomalacia, cerebral cortex, and nucleus of brainstem	None
28 Mar	Nursery 11	4	Multifocal polioencephalomalacia, cerebral cortex	None
30 Mar	Sow Farm 3	3	None	None
30 Mar	Nursery 12	6	None	Pulmonary edema; diffuse hepatic vacuolization
4 Apr	Wean-to-finish 3	3	Neuronal necrosis and polioen- cephalomalacia, cerebral cortex	Atrophic enteritis, interstitial pneumonia
4 Apr	Nursery 13	6	None	Pulmonary edema, suppurative bronchointerstitial pneumonia, necrotizing bronchiolitis
18 Mar	Nursery 14	4	Mononuclear cells, meninges of the brain	Interstitial pneumonia, glomerulonephritis

^{*} Neurologic signs were observed 5 to 7 days after pigs were weaned at approximately 19 days of age in several nursery farms in North Carolina. The table does not include findings from the College of Veterinary Medicine, North Carolina State University.

Figure 4: Neurologic signs were observed in pigs 5 to 7 days after weaning in several nursery farms in North Carolina. Within 2 months, the clinical signs were observed in pigs prior to weaning. Figures 4 to 9 provide the descriptions of histopathology associated with pigs (17 days of age) submitted to the College of Veterinary Medicine, North Carolina State University. Cerebral cortex; Pig 5. The left half of the image exhibits an extensive area of laminar, neuronal necrosis (N) and neuropil rarefaction due to edema with a relatively abrupt transition to viable cerebral cortex (V) on the right side of the image. There is a mild, mononuclear infiltrate within the overlying meninges. Hematoxylin and eosin, 4× magnification.

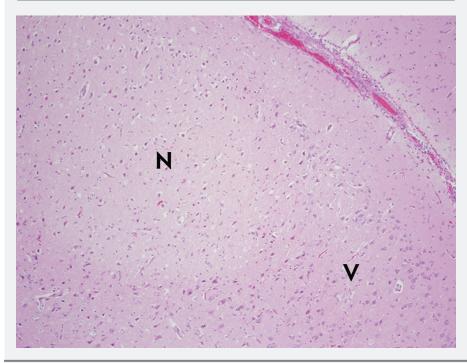
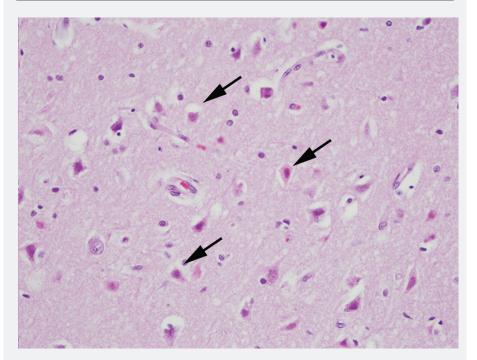


Figure 5: Cerebral cortex; Pig 5. Higher magnification of neuronal necrosis shown in Figure 4. The necrotic neurons are shrunken and angular with hypereosinophilic cytoplasm and pyknotic nuclei (arrows). Vessels in the section are lined by hypertrophied endothelial cells. Necrotic neurons and vessels are surrounded by edema. Hematoxylin and eosin, $40 \times$ magnification.



cosis in ruminants involves metabolism of ingested sulfur compounds to hydrogen sulfide gas by ruminal microbes. This toxic gas is either absorbed though the ruminal wall or eructated and inhaled. Hydrogen sulfide is thought to inhibit cytochrome oxidase and interfere with aerobic metabolism in the brain. However, it also may be involved in the formation of free radicals or act as an exogenous neuromodulator. 16 It was unfortunate that sulfur content in the diet or drinking water was not evaluated in the present case. Attempts to induce neuronal lesions in 10-week-old, 7- to 14-kg pigs through thiamine-deficient diets historically have been unsuccessful. Experimentally induced thiamine-deficiency in pigs caused cardiac dilation and hypertrophy with myocardial necrosis.7 These histological lesions were not routinely evident in the pigs showing clinical signs and submitted to the various diagnostic laboratories.

On the basis of the positive response to supplemental thiamine injections, it was suspected that the thiamine in the diet was either destroyed or unavailable. Thus, additional thiamine was added to the vitamin premix. Animals do not synthesize thiamine and obtain daily requirements from their diets, storing excesses in the liver. Metabolic disorders are associated with thiamine deficiency; among the attendant clinical signs are diarrhea, reduced growth rate, weight loss, and anorexia, while central nervous system disorders usually manifest with severe depletion at a later stage of thiamine deficiency. It is speculated that the apoenzyme-thiamine complexes in the brain, which protect the brain tissue from sudden changes in enzyme activity, are responsible for this clinical presentation. Body reserves are reported to be sufficient for only 3 weeks in ruminants, but are not documented for swine.¹⁷ Even though thiamine is active in most cells, cells of the nervous system and heart seem particularly sensitive to the effects of thiamine deficiency. 18

The clinical presentation of thiamine deficiency in humans is similar to the clinical signs noted in the present case. Thiamine deficiency is not uncommon in human populations in the Far East, where infants suckling from thiamine-deficient mothers develop clinical signs of beriberi syndrome and slow growth rates. ¹⁹ Beriberi refers to the lack of thiamine pyrophosphate, which is the active form of thiamine in humans. Thiamine stores are usually depleted within

1 month, and clinical signs, beginning as early as 1 week after the last ingestion, include weakness and peripheral neuropathy. The "dry" form of beriberi refers to the neurologic disease where individuals have impairment of sensory, motor, and reflex functions of the extremities due to degeneration of myelin in the muscular sheaths.²⁰

The "wet" form of beriberi refers to a thiamine deficiency with cardiovascular involvement, where peripheral vasodilation occurs, causing high cardiac output. This initiates water and sodium retention through the renin-angiotensin-aldosterone system, which ultimately results in fluid overload, peripheral edema, and myocardial injury. If this occurs acutely, there can be fatal fulminant cardiac failure. This cardiovascular involvement possibly contributed to the pathogenesis of severe pulmonary edema and sudden death observed in the older population of pigs in the present case.

With the presentation of periweaning-age pigs being affected, it was likely that inadequate thiamine was transferred through the milk. In one study, ¹⁹ rats were fed a thiamine-deficient diet during the latter stages of gestation and the entire lactation phase. When the dams were fed the thiaminedeficient diet, transfer of thiamine from the dam to the pups was reduced after day 12 of lactation. Extrapolating to the present case, the sows' stores of thiamine became depleted as lactation progressed, and the piglets no longer received adequate thiamine in the milk. Presumably, the sows were not affected because reserves of thiamine were greater. An early study successfully induced thiamine deficiency in young pigs.²¹ Pigs were taken from sows at 3 or 4 days of age and fed a thiamine-depleted diet. Pigs appeared clinically normal for the initial 12 days, but became weak, started vomiting, and died within 4 weeks. Heart lesions were noted in thiamine-deficient pigs, but not in pigs that were necropsied after supplementation of thiamine.²¹ These observations confirm the time frame of clinical disease observed in the periweaning pig population in the present case; however, myocardial lesions were not observed in this case. Rather, the neurological lesions were consistent with the clinical signs.

The authors do not recommend routine supplementation of diets with thiamine, nor do we believe that thiamine injections are required under normal circumstances on commercial farms. In the present case, com-

Figure 6: Cerebral cortex; Pig 5. Higher magnification of viable cortex shown in Figure 4. There is mild gliosis characterized by increased numbers of enlarged astrocytes (arrows) with vesicular nuclei that are occasionally present in pairs (Alzheimer's type II astrocytes). Hematoxylin and eosin, 40× magnification.

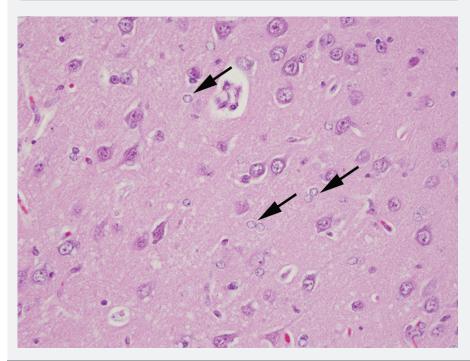
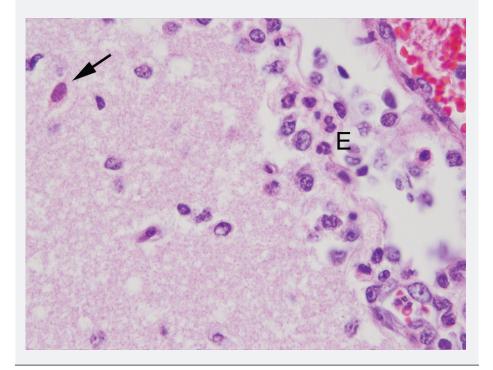


Figure 7: Cerebral cortex; Pig 8. High magnification of meningeal infiltrate overlying a region of cortical necrosis reveals small numbers of eosinophils (E) in addition to the mononuclear cells seen in most of the affected pigs. A necrotic neuron (arrow) is present in the top left of the field. Hematoxylin and eosin, 100×100 magnification.



promised dietary thiamine levels during feed manufacturing contributed to the PEM.

Implications

- Pigs may develop histological lesions similar to those reported in ruminants affected with PEM.
- In this case, the causation diagnosis
 of thiamine destruction in the feed
 is speculative; however, practitioners
 need to consider thiamine deficiency
 as a potential cause of neurological
 disorders in young pigs.

Conflict of interest

None reported.

Disclaimer

Scientific manuscripts published in the *Journal of Swine Health and Production* are peer reviewed. However, information on medications, feed, and management techniques may be specific to the research or commercial situation presented in the manuscript. It is the responsibility of the reader to use information responsibly and in accordance with the rules and regulations governing research or the practice of veterinary medicine in their country or region.

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Figure 8: Brainstem; Pig 2. There is a focus of neuronal necrosis (N), neuropil vacuolation, and mild, mononuclear cell perivascular cuffing (arrow) within the periventricular grey matter. Hematoxylin and eosin, 20× magnification.

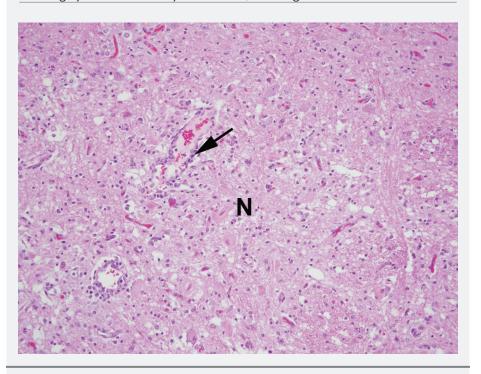
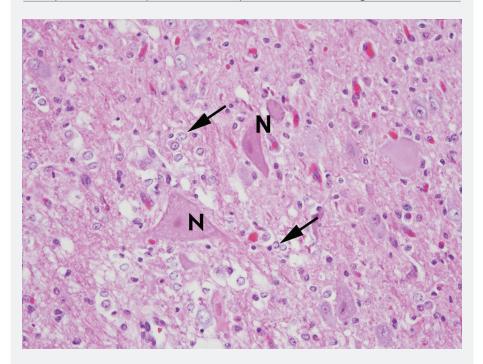


Figure 9: Brainstem; Pig 2. Higher magnification of neuronal necrosis (N) and neuropil vacuolation shown in Figure 8. Additionally, several Alzheimer's type II astrocytes (arrows) are present. Hematoxylin and eosin, 40× magnification.



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News from the National Pork Board **Pork** Checkoff.



Checkoff research delivers predictable return of more than two to one

In 2014, pork producers continued to battle porcine epidemic diarrhea virus and faced other challenges, but as always, the investment they've made through the Pork Checkoff into science- and technology-based research continued to deliver dividends.

"Our long-term focus on research, education, and the sharing of information is paying off," said Dale Norton, National Pork Board president and producer from Bronson, Michigan.

"The ongoing investment in Checkoff funds over the year has provided us with solutions that help contribute to a stronger pork industry."

From a financial perspective, Checkoff funds allocated to research in science and technology don't simply work alone. They actually draw in outside funding from other sources such as land-grant universities and allied industry. This is shown in a 2014 review of

all funded National Pork Board science and technology projects from 2007 to 2011, which found that for every dollar of Checkoff investment, more than two additional dollars are drawn from outside sources to help find solutions to mutual challenges that are facing the pork industry.

For more information, contact Paul Sundberg at PSundberg@pork.org or 515-223-2764.

Updates from Checkoff's science and technology committees

At the National Pork Board's recent Unified Research Meeting held in Orlando, Florida, each of the committees in the science and technology area gave updates and conducted related business. Here are some selected highlights.

Animal science

The committee invested a substantial amount of time discussing their role in a successful 2015 Strategic Plan by 2020. The committee was asked to evaluate their role in each of the objectives under each of the three goals: build consumer trust, drive sustainable production, and grow consumer demand.

Research update (Sow Lifetime Productivity). A summary of the preliminary report was given on how research is being conducted and collaborated upon. An additional project was discussed, and the committee agreed with the Sow Lifetime Productivity Scientific Working Group that this project should be funded.

Review of research proposals (High Feed Cost Mitigation Research). A total of 20 research proposals were submitted to the High Feed Cost Mitigation RFP. The committee voted to exclude seven proposals from farther consideration on the basis of poor scientific score and producer review.

Animal welfare

The committee received an update on the progress of the Industry Audit Task Force and the newly introduced Common Swine Industry Audit. The committee discussed

the future of the third-party verification component of PQA Plus now that the common audit is available. A motion was made to reallocate funds originally budgeted for PQA Plus third-party verification and use them to build a dashboard for aggregating data from the common audit. The motion was seconded and was approved by voice vote.

Checkoff staff presented proposed content for the animal-care chapter of the next PQA Plus version. Committee reviewed the chapter and offered revisions to the content. Staff was instructed to incorporate the edits and recirculate for final approval of the content.

An Iowa State University researcher presented mid-project data from a Checkofffunded research study focusing on fitness to transport of pigs arriving at consolidation markets.

Producer/Public Health and Workplace Safety Committee

Jim Lummus, Checkoff's retiring director of producer learning and development, introduced Karen Hoare as his successor.

The Safe Pig Handling training materials, revision of the Employee Safety Tool Kit, and the Workplace Safety Assessment research were completed and distributed.

A half-day session was held on antimicrobial resistance and benchmarking for the industry. Several speakers were invited to provide an overview of their activities and perspectives. Invited speakers and topics

included Dennis Treacy, EVP and Chief Sustainability Officer, Smithfield, speaking on "PCAST report on combating antibiotic resistance," followed by Craig Lewis, Veterinary Medical Officer Center for Veterinary Medicine, FDA, on "FDA antimicrobial resistance strategy," and finally, Dr Peter Davies, Professor of Swine Health, University of Minnesota.

Environment committee

Allan Stokes, Checkoff's director of environment, gave an update on the National Pork Board's Sustainable Pork Framework and associated sustainability efforts, including the Four Pillars of Environmental Sustainability. The committee members then discussed how the National Pork Board can ensure the Sustainable Pork Framework components are implemented and progress in sustainability efforts continues.

Karen Hoare, director of producer learning and development with the National Pork Board, facilitated a committee discussion on including environmental components in National Pork Board educational programs and solicited member input to identify key topics to be trained, identify sources of core materials, and identify persons who might serve on a working group to assist in developing the training materials.

Pork safety, quality, and human nutrition

The committee considered seven human nutrition proposals and funded two. They considered one pork-safety proposal and funded it, while they also considered three pork-quality research proposals, but decided they needed additional input before deciding. In an open discussion, the committee established food-safety baseline studies for the National Pork Board's strategic plan.

Other business centered on the impact of heavy pigs on pork safety and a study on lymph nodes. In addition, three researchers have submitted a joint quality proposal to consider for funding.

Related to nutrition, the committee discussed human health effects of inclusion of pork in diets, cardiovascular disease, and adult gut health, along with other health topics.

Swine health

The committee provided comments regarding the proposed concept paper on the National List of Reportable Animal Diseases. Comments will go to USDA:

Reviewed an issue brief on feral swine surveillance and developed a committee position statement to be used for comments back to USDA Veterinary Services and USDA Wildlife Services regarding future disease surveillance in feral swine; Provided comments on the proposed changes in pseudorabies virus/swine brucellosis surveillance. Comments will go to USDA; and

Discussed the direction that the current swine enteric coronavirus diseases plan should take: continue, discontinue, or adjust the program.

America's Pig Farmer of the Year award applications due May 15

The National Pork Board's new America's Pig Farmer of the Year award is accepting applications until May 15. The award will honor the US pork producer who best excels at raising pigs using the We Care ethical principles and wants to share with the public how he or she does that. The program builds on many elements behind the successful 20-year run of the now-retired Environmental Stewards Award program.

"The public is the main audience rather than our own industry because that's who has questions about how we raise pigs," said Brad Greenway, vice president of the National Pork Board and chairman of the Stewards Task Force, which oversaw creation of the new program. "Producers demonstrate the We Care ethical principles on their farms every day, and the new award is a unique way to share that with the public," he said.

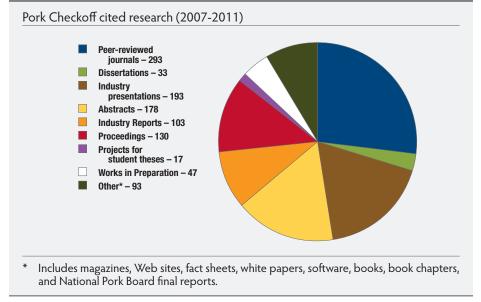
The intent is to establish the winner as a practical expert in pig handling and pork production, according to Kevin Waetke, vice president of strategic communications for the Pork Checkoff. "Consistent with the National Pork Board's new strategic plan, we want to build consumer trust through on-farm transparency and accountability," he said. "The focus is on environmental sustainability, along with animal welfare, production efficiency, the adoption of best practices, and a commitment to continuous improvement."



For more information go to www. americaspigfarmer.com or contact Mike King at MKing@pork.org or 515-223-3532.

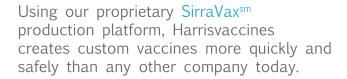
Checkoff research earns multiple citations

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AASV NEWS

AASV announces 2015 officers

Dr Ron Brodersen was installed as the president of the American Association of Swine Veterinarians at the association's 46th annual meeting in Orlando, Florida. He succeeds Dr Michelle Sprague, who is now immediate Past President. Dr George Charbonneau has ascended to President-elect. The newly elected Vice President is Dr Alejandro "Alex" Ramirez.

AASV President

Dr Ron Brodersen (ISU '79) grew up on a livestock farm near Coleridge, Nebraska. He attended the University of Nebraska-Lincoln and Iowa State University where he received a DVM degree, and also attended the University of Illinois EVP program. Dr Brodersen has been providing swine veterinary services in Hartington, Nebraska, since 1990. His veterinary practice recently became a part of Suidae Health & Production. He also owns Whole Hog Genetics. He was active on the Nebraska Pseudorabies Eradication Task Force in the 1990s. Dr Brodersen has been active in the AASV, serving on the board of directors as well as the pharmaceutical and boar stud committees. He has also served as chairman of the AASV Foundation. The AASV recognized him as the Swine Practitioner of the Year in 2003.

When asked to comment on his thoughts about the future of AASV and his tenure as president, Dr Brodersen said, "I am anxiously looking forward to serving as president of the American Association of Swine Veterinarians. I plan to continue promoting our members as professional specialists of swine health and equal specialists of swine welfare. Also, our newly updated mission statement expands our role beyond educating veterinarians to include advocacy of swine industry issues. I expect this will be an interesting and exciting year!"

AASV President-elect

Dr George Charbonneau (ON '81) grew up in Arnprior, Ontario. He obtained his Doctor of Veterinary Medicine from the Ontario Veterinary College and established



AASV officers (left to right) Dr Ron Brodersen, Dr George Charbonneau, Dr Alex Ramirez, and Dr Michelle Sprague

Photo courtesy of Tina Smith

a veterinary practice serving southwestern Ontario. George is currently a partner in South West Ontario Veterinary Services and is based in Stratford, Ontario. Dr Charbonneau has been very active in the Canadian swine industry. He has served as the president of the Canadian Association of Swine Veterinarians, Ontario Association of Swine Veterinarians, and the Ontario Pork Congress. He was involved in the formation of, and served as the initial chairman of, the Ontario Pork Industry Council. He also represented Canadian swine veterinarians as a district representative on the board of directors of the American Association of Swine Veterinarians. He was the 2012 recipient of the AASV Swine Practitioner of the Year award.

AASV Vice President Dr Alejandro "Alex" Ramirez (ISU '93)

grew up in Guadalajara, Mexico. He obtained his Doctor of Veterinary Medicine degree from the Iowa State University (ISU) College of Veterinary Medicine and joined Valley Veterinary Center, a mixed-animal practice, in Cherokee, Iowa. In 2004, Alex left practice and returned to ISU to pursue a teaching career. He obtained a Master of Public Health degree from the University of Iowa and concluded a PhD at ISU in 2011.

Dr Ramirez joined AASV in 2002. He first served as a substitute judge for the student presentations at the AASV Annual Meeting. Shortly thereafter he was asked to co-chair the student oral competitions. He has also co-chaired the Collegiate Activities Committee for the past few years and has served on the *Journal of Swine Health and Production* Editorial Board since 2010. He has represented District 6 on the AASV Board of Directors since 2013.

"It is an honor and a privilege to be able to serve our great association as vice president. I am hopeful that I can continue providing the great leadership that has already been provided by all those officers whom I follow," he noted following his election.

AASV Past President

Dr Michelle Sprague (ISU '05) grew up on a small farrow-to-finish and row-crop farm in Glenwood, Iowa. Following graduation from the Iowa State University College of Veterinary Medicine in 2005, she joined the Audubon Manning Veterinary Clinic (AMVC) in Audubon, Iowa. She is currently a partner and director of sow health at AMVC. Her responsibilities include overseeing animal health, biosecurity, food safety, and animal welfare on all the clinic's managed sow farms.

Operation Main Street Training

Co-sponsored by AASV and the National Pork Board

Members of AASV like Drs Rick Tubbs, Craig Rowles, Amy Woods, Jeff Harker, Peggy Anne Hawkins, and Gene Nemechek – to name just a few of the 86 veterinarians participating in Operation Main Street – are making a difference by sharing the facts about pig care and pork production with veterinary students, dieticians, and civic groups across the United States.

You can join your colleagues in the effort to counter misunderstanding and misinformation about the swine industry by becoming a trained Operation Main Street (OMS) speaker. Two OMS speaker-training opportunities will be held during World Pork Expo in Des Moines, Iowa: June 2-3, June 4-5.

In 2011, AASV and the National Pork Board partnered to train veterinarians as OMS speakers with a goal to schedule a speaker in all 30 schools of veterinary medicine. To date, trained veterinarians have presented at 26 of 30 schools, reaching more than 5000 students through this program.

The training updates participants on what activists are saying about agriculture today

and provides attendees with the needed tools and presentations to address those concerns in a science-based, proactive manner. The objective is to equip veterinarians to speak to veterinary students and professional groups, including dieticians. Any AASV member interested in becoming a trained OMS speaker and helping in this endeavor is invited to participate.

For more information, contact MaryWonders at the National Pork Board (Tel: 515-223-3535; E-mail: Mwonders@pork.org).

Call for abstracts – AASV 2016 Student Seminar

Veterinary Student Scholarships

The American Association of Swine Veterinarians announces an opportunity for veterinary students to make a scientific presentation during the Student Seminar at the AASV Annual Meeting in New Orleans, Louisiana, on Sunday, February 28, 2016. Interested students are invited to submit a 1-page abstract of a research paper, clinical case study, or literature review for consideration. The submitting student must be a current (2015-2016) student member of the AASV at the time of submission, and must not have graduated from veterinary school prior to February 28, 2016. Submissions are limited to one (1) abstract per student.

Abstracts and supplementary materials must be received by Dr Alex Ramirez (alex@ aasv.org) by 11:59 PM Central Daylight Time on Monday, September 21, 2015 (firm deadline). All material must be submitted electronically. Late abstracts will not be considered. You should receive an e-mail confirming the receipt of your submission. If you do not receive this confirmation e-mail, you must contact Dr Alex Ramirez (alex@ aasv.org) by Wednesday September 23, 2015, with supporting evidence that the submission was made in time, otherwise

your submission will not be considered for judging. The abstracts will be reviewed by an unbiased professional panel consisting of a private practitioner, an academician, and an industry veterinarian. Fifteen abstracts will be selected for oral presentation in the Student Seminar at the AASV Annual Meeting. Students whose papers are selected will be notified by October 15, 2015, and will be expected to provide the complete paper or abstract, reformatted for publication, by November 16, 2015.

As sponsor of the Student Seminar, Zoetis provides a total of \$20,000 in support to fund travel stipends and the top student presenter scholarship. The student presenter of each paper selected for oral presentation receives a \$750 stipend to help defray the costs of attending the AASV meeting.

Each veterinary student whose paper is selected for oral presentation competes for one of several veterinary student scholarships awarded through the AASV Foundation. The oral presentations will be judged to determine the amount of the scholarship awarded. Zoetis funds the \$5000 scholarship for the student whose paper, oral presentation, and supporting information are judged

best overall. Elanco Animal Health provides \$20,000 in additional funding, enabling the AASV Foundation to award \$2500 each for $2^{\rm nd}$ through $5^{\rm th}$ place, \$1500 each for $6^{\rm th}$ through $10^{\rm th}$ place, and \$500 each for $11^{\rm th}$ through $15^{\rm th}$ place.

Abstracts that are not selected for oral presentation in the Student Seminar will be considered for participation in a poster session at the annual meeting. Zoetis and the AASV fund a stipend of \$250 for each student who is selected and participates in the poster presentation. In addition, the presenters of the top 15 poster abstracts compete for awards ranging from \$200 to \$500 in the Veterinary Student Poster Competition sponsored by Newport Laboratories.

Complete information for preparing and submitting abstracts is available on the AASV Web site at www.aasv.org/annmtg/2016/studentseminar.htm. Please note: the rules for submission should be followed carefully. For more information, contact the AASV office (Tel: 515-465-5255; Fax: 515-465-3832; E-mail: aasv0 aasv.org).

AASV Proceedings Archive online

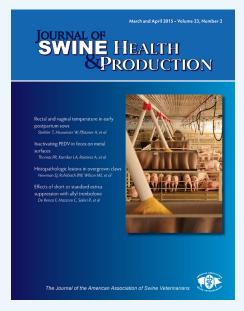
With the successful transition to electroniconly proceedings for the 2015 AASV Annual Meeting, AASV members won't be receiving the conference proceedings in the mail this year. Instead, members will find the familiar "big book" available as a single PDF – as well as PDF files for each of the preconference seminar booklets – in the newly created online AASV Proceedings Archive. To download the files, visit https://www. aasv.org/library/proceedings/ or look under the "Resources" menu tab on the AASV Web site for "AASV Meeting Proceedings."

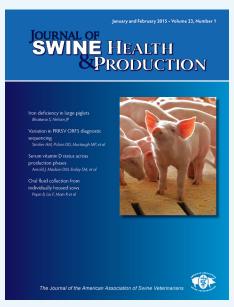
You'll want to make sure your AASV membership has been renewed for 2015, and you'll need your AASV member username and password: If they're not handy, contact the AASV office or use the "Reset Password" link in the upper right of the AASV web site (https://www.aasv.org) to have them e-mailed to you.

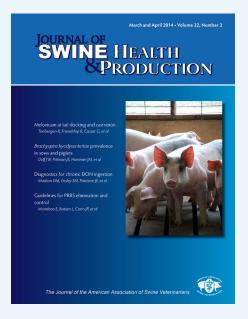
As in the past, PDFs for each of the individual proceedings papers will continue to be available as part of the AASV Swine Information Library, https://www.aasv.org/library/swineinfo/. This fully-searchable, online library of more than 12,000 proceedings papers and journal articles is just one of the many benefits enjoyed by AASV members.

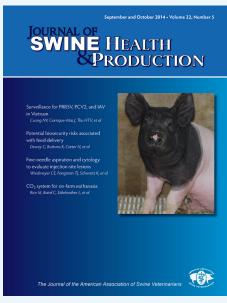


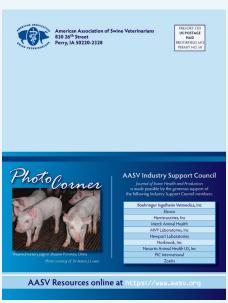
SUBMIT YOUR PHOTOS

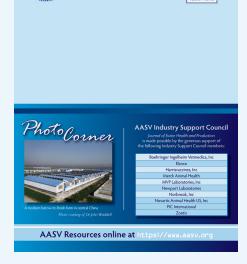












The Journal of Swine Health and Production would like to publish digital photographs submitted by our readers. Images used either on the front cover or in the photo corner on the back cover are to represent healthy pigs and modern production facilities. Please ensure that the photos do not include people. Select the largest image size available on your camera, of the quality or compression that allows you to store the fewest images on a given memory card. Do not resize, crop, rotate, or color-correct the image prior to submission to the journal. Please send the images by e-mail attachment to tinalaasv.org. Tina will also need to know your name, affiliation, and the approximate location of the subject, or other details that you would like to submit that describe the image.

ANNUAL MEETING REPORT

2015 AASV Annual Meeting sets records

The American Association of Swine Veterinarians (AASV) held its 46th annual meeting in Orlando, Florida, February 28 to March 3, 2015. The meeting, held at the Buena Vista Palace Hotel & Spa, drew record attendance of 1109 total attendees including 654 paid registrants (also a record), 285 international members, and 138 students. The participants represented 23 countries, with 25% of attendees from outside the United States. Total attendance included 240 allied industry representatives from 84 exhibitors, manning a record 85 technical tables. The students in attendance represented 28 veterinary schools!

The meeting participants attended numerous educational sessions, including 11 preconference workshops, two general sessions, three break-out sessions, research topics, industrial partner sessions, 15 student seminar presentations, and 72 posters.

Dr Greg Stevenson opened the Monday General Session with the Howard Dunne Memorial Lecture. His presentation, entitled "Because it's the right thing to do" reminded the audience of the importance of maintaining your integrity and making sure you "like who you see in the mirror." He concluded by saying, "There is much at stake for each of us as individuals, for our profession, and for our organization. Choose integrity, because it is the right thing to do."

Dr Scanlon Daniels presented the Alex Hogg Memorial Lecture entitled "Influence and advocacy: Opportunities for swine veterinarians." He challenged the attendees to consider how swine veterinarians can re-establish their relationship with society. The answer, he noted, "is balancing facts with values."

The second half of the Monday morning session focused on porcine epidemic diarrhea virus, coronavirus immunity, and clinical presentation. The Tuesday General Session addressed the issues associated with the introduction of transboundary and foreign animal diseases. All of the General Session presentations on Monday and Tuesday were video recorded and will be posted in the video library of the AASV Web site in the near future.

The AASV Awards Reception was held Monday night, followed by the AASV Foundation's annual fund-raising auction. Dr Randy Jones, 2011 AASV President and chair of the 2015 Awards Selection Committee, presented the recipients of the Howard Dunne Memorial Award (Dr Butch Baker), the Technical Services/Allied Industry Veterinarian of the Year Award (Dr Kerry Keffaber), the Young Swine Veterinarian of the Year Award (Dr Megan Inskeep), the Meritorious Service Award (Dr Howard Hill), and the award for Swine Practitioner of the Year (Dr Larry Coleman).

In addition, 15 AASV committees met during the annual meeting. The 2015 officers, Drs Ron Brodersen, President; George Charbonneau, President-elect; Alex Ramirez, Vice President; and Michelle Sprague, Past President, were introduced during the Annual Business Meeting on Tuesday morning. The board also welcomed incoming representatives: District 1, Dr Lynette Holman (re-elected) and District 7, Dr Scanlon Daniels.

If you would like to provide feedback on this year's meeting or suggestions for future meetings, please complete the short online survey at http://fluidsurveys.com/s/AASVZØ15. The 2016 annual meeting will be held February 27 to March 1 in New Orleans, Louisiana.

Swine Practitioner of the Year

Dr Larry Coleman was named **2015 Swine Practitioner of the Year** by the American Association of Swine Veterinarians (AASV). The award is given to the swine practitioner who has demonstrated an unusual degree of proficiency and effectiveness in the delivery of veterinary service to clients.

Dr Coleman obtained his Doctor of Veterinary Medicine degree from the University of Missouri Veterinary College in 1980. Following graduation, he accepted a position at a mixed food-animal practice in Broken Bow, Nebraska. In 1986, Dr Coleman left Broken Bow to spend a year at the North Carolina State University working with swine integrators in that state. He returned to Broken Bow in 1987 and opened his own food-animal



Dr Larry Coleman, recipient of the AASV Practitioner of the Year Award

practice, where he remains to this day, having been joined by Dr Russ Rice and Dr Clayton Smith. Dr Coleman's personal veterinarymedicine passion is the "art and science" of getting employees to emotionally engage when they are taking care of animals.

Dr Coleman joined AASV in 1984 and has served on various AASV committees since that time.

Asked to comment about receiving this award, Dr Coleman replied, "I am very honoured to have received the Swine Practitioner of the Year Award. My involvement with the AASV and its members has been a great aid in the development of my professional skills, as well as establishing many valuable business and personal friendships."

Dr Coleman is married to Renea Coleman and they reside in Broken Bow, Nebraska. They have two grown daughters: Kinsi, who is a high school teacher in Alma, Nebraska, and Kia, who is a second-year student at Hastings College in Hastings, Nebraska, with plans to become a veterinarian.

Howard Dunne Memorial Award

Dr Rodney "Butch" Baker received the American Association of Swine Veterinarians (AASV) **2015 Howard Dunne Memorial Award** during the association's 46th annual meeting March 2 in Orlando, Florida. The



Dr Rodney (Butch) Baker, recipient of the Howard Dunne Memorial Award

award recognizes an AASV member who has made important contributions and provided outstanding service to the association and the swine industry.

Dr Baker was raised on a small, diversified farm in Owensboro, Kentucky, where he gained an early appreciation for farm life, livestock, and veterinary medicine. He received his DVM from the Auburn University College of Veterinary Medicine in 1978 and earned a master's degree in veterinary diagnostics and production-animal medicine from Iowa State University (ISU) in 1999.

Dr Baker joined ISU in 2006 as a senior clinician in the College of Veterinary Medicine. Previously, Baker served as partner in a multi-veterinarian multi-location practice primarily involved in food-animal practice. He served as an Area Veterinary Consultant for the Pig Improvement Company (PIC) and numerous other producers during the transformation of the US pork industry to the modern structure of today. After leaving practice he worked for Bayer Animal Health, PIC, Premium Standard Farms, and North Carolina State University. Dr Baker also recently served as the interim director of the Iowa Pork Industry Center at ISU. He is part owner of a 2400-sow breed-to-wean farm in Georgia and another farm in Kentucky that is dedicated to sow well-being research and leased to Cargill Meat Solutions.

Dr Baker became a member of AASV in 1978 and was president of the association in 2009. He has served on numerous AASV issue-based committees and AASV program planning committees, as well as the AASV Foundation Auction Committee and AASV Foundation Board. He is also an AASV Foundation Heritage Fellow. In 2012, he was honored as the Agriculture Alumnus of the Year by Western Kentucky University's Ogden College of Science and Technology. He was recognized as a Master of the Pork Industry by National Hog Farmer magazine in 2013, and received the Honorary Master Pork Producer plaque at the 43rd annual Iowa Pork Congress on February 1, 2015.

When asked what it meant to him to receive the Howard Dunne Memorial Award he responded, "I am truly honored and humbled by this unexpected surprise. It is certainly the most appreciated recognition of my long career. I thank the AASV awards committee for choosing me as the recipient."

Butch and his wife, Emma, currently reside in Ames, Iowa. They have three children (Brad, Amy, and Will) and three grandchildren (Marissa, Paige, and Reith).

Meritorious Service Award

Dr Howard Hill was named the 2015 recipient of the American Association of Swine Veterinarians (AASV) **Meritorious Service Award.** The award recognizes individuals who have provided outstanding service to the AASV.

A native of southern California, Dr Hill earned his DVM degree from the University of California-Davis in 1969. Hill spent a year in private practice in Vista, California, before deciding to pursue his master's degree (1972) and PhD (1974) in veterinary microbiology from Iowa State University.

Dr Hill retired as the director of animal well-being for Iowa Select Farms at the end of 2012, but has been retained as an advisor with a focus on animal well-being. Hill began his career with Iowa Select Farms in 2000, when he joined the company as director of production. In 2001, he was promoted to chief operating officer and continued in that role until 2009.

Hill served as director of veterinary services and multiplication for Murphy Family Farms (1994-2000) in Rose Hill, North Carolina. Previously, Hill served as head of veterinary microbiology in the Iowa State University Veterinary Diagnostic Laboratory (1974-1994).

Dr Hill joined AASV in 1979. He has served on AASV program planning committees and as AASV president in 1996. In addition, he received the Howard Dunne



Dr Howard Hill, recipient of the Meritorious Service Award

Memorial Award in 1992 and is an AASV Foundation Leman Fellow. He received Iowa State University's Science with Practice Award in 2011. Hill was one of nine veterinarians appointed to serve on the US Department of Agriculture's Advisory Committee on Animal Health (2011-2013). He is a member of the Story County Pork Producers and serves on the board of directors of the Iowa Pork Producers Association. He is the immediate past president of the National Pork Producers Council.

When asked to comment about receiving the award, Hill responded, "I am very honored and humbled to receive this award from the AASV. We all belong to several organizations, but I have always felt like the AASV has been my "home organization." There is no other professional organization that does more for its members and future members (students) than AASV. It is an organization of members helping members. The strength of the organization has been the leadership of those members who have given freely of their time and the guidance from an excellent staff. Thank you AASV for this award."

Dr Hill and his wife, Nancy, reside in Cambridge, Iowa. They have three children (Allison, Eric, and Jared) and 10 grandchildren. Their livestock business includes hogs and cattle. Hill and his son also farm 2600 acres of corn, soybeans, and alfalfa.

Young Swine Veterinarian of the Year Award

The American Association of Swine Veterinarians (AASV) **Young Swine Veterinarian of the Year Award** was presented to **Dr Megan Inskeep** during the 46th annual meeting of the AASV in Orlando, Florida. It is given annually to an AASV member 5 or fewer years post graduation who has demonstrated the ideals of exemplary service and proficiency early in his or her career.

Dr Inskeep grew up in Wilson, North Carolina, where her parents, Gene and Susan Nemechek, still reside. Exposure to farm life came at a very early age for Dr Inskeep. She developed an interest in animals beginning on her grandparents' dairy farm, visiting pig farms with her veterinarian dad, and working in a small-animal clinic during high school. These experiences sparked her interest in becoming a veterinarian. She worked two summers on sow farms gaining experience in farrowing and gestation barns.

Majoring in animal science as an undergraduate at North Carolina State University (NCSU), she was active in numerous clubs and organizations. She was among the first class of Food Animal Scholars accepted into the NCSU College of Veterinary Medicine. She earned her DVM in 2010. She participated in the National Pork Board's Operation Main Street as a student presenter and continues to participate as a practitioner. To date, she has given more than 30 presentations to civics groups and high schools in her area.

She was active all 4 years in the swine club and the bovine club at the veterinary school. She served as president of the swine club her



Dr Megan Inskeep, recipient of the Young Swine Veterinarian of the Year Award

junior year, organizing "pig pickin" lunch fund raisers and program speakers for the club meetings. She says that she will never forget the generosity of the veterinarians and their families and the lifelong friendships she made during numerous veterinary internships at swine practices throughout the Midwest. She continues that spirit of generosity by hosting numerous veterinary students and offering them the opportunity to experience swine veterinary practice and the modern swine industry. She received the Swine Proficiency Award at the conclusion of her senior year of veterinary school.

"I am very honored to receive this award, and feel very fortunate to be a part of such a remarkable organization and industry. I can't begin to give enough thanks to my family and mentors over the years for their support, and hope that I can give back as much as they have given to me," commented Dr Inskeep.

Currently, she is a veterinarian at Rensselaer Swine Services with Dr Tom Gillespie. She also works with Dr Chuck Hannon and Donor Solutions, Inc, specializing in cattle donor and reproductive services. She lives in Rensselaer, Indiana, with her husband Bryan Inskeep, and they are expecting their first child in August.

Technical Services/Allied Industry Veterinarian of the Year Award

Dr Kerry Keffaber received the American Association of Swine Veterinarians (AASV) Technical Services/Allied Industry Veterinarian of the Year Award. Established in 2008, the award recognizes swine industry veterinarians who have demonstrated an unusual degree of proficiency and effectiveness in delivery of veterinary service to their companies and their clients, as well as given tirelessly in service to the AASV and the swine industry.

Dr Keffaber was recognized for his years in technical service at Elanco Animal Health. Since joining Elanco in 2002 as a Swine Technical Services Consultant, he has served numerous roles including Director of Technical Consulting in the Swine Business Unit and Director of Swine Innovation. His current title is Advisor Scientific Affairs & Policy. In this role, he leads global and US efforts to help others in regulatory, government, and the food-supply chain stay informed and establish policy on current global animal



Dr Kerry Keffaber, recipient of the Technical Services/Allied Industry Veterinarian of the Year Award

health, scientific research, and food-safety topics. In addition, he participated in the first Elanco-sponsored study tour to Cameroon, Africa, with Heifer International with a group of swine practitioners.

Dr Keffaber received his BS and DVM (1981) from Purdue University. Following graduation, Dr Keffaber joined the Manchester Veterinary Clinic where he focused on large animals and swine. In 1987, he founded the Swine Health Center in Roann, Indiana, where he practiced until joining Elanco Animal Health. Dr Duane Long continued the practice.

Active in the AASV since 1981, Dr Keffaber served as a district director for two terms and as president of the association in 2008. In addition, he has chaired the AASV PRRS subcommittee and participated on several planning committees for the association's annual program. He was also a graduate of the inaugural Executive Veterinary Program in Swine Health Management class at the University of Illinois.

When asked to comment on what the award meant to him, Dr Keffaber said, "AASV is filled with quality people that are excellent scientists and great examples of leadership and integrity; to be recognized with this honor is humbling and quite a nice surprise and serves as a challenge to continue to work to help animals and people."

Dr Keffaber and his wife, Betsy, reside in Fishers, Indiana. They have three children (Brad, Megan, and Abbey) and four grandchildren, with another on the way.

AASV Foundation announces student scholarships

The American Association of Swine Veterinarians (AASV) Foundation awarded scholarships totaling \$25,000 to 15 veterinary students during the 46th AASV Annual Meeting in Orlando, Florida.

Joseph Thomas, Iowa State University, received the \$5000 scholarship for top student presentation. His presentation was titled "Effect of porcine epidemic diarrhea virus infectious doses on outcome of infection in naive neonatal piglets and weaned pigs." Zoetis provided the financial support for the top student presenter award.

Additional scholarships totaling \$20,000 were funded by Elanco Animal Health. Four veterinary student presenters received \$2500 scholarships: **Brianna Peters**, University of Tennessee; **Christopher Sievers**, Iowa State University; **Alyssa Taplett**, Iowa State University; and **Kathleen Wood**, North Carolina State University.

Five veterinary student presenters received \$1500 scholarships: **Hunter Baldry**, University of Minnesota; **Brigitte Mason**, University of Illinois; **Jacqueline Myers**, Iowa State University; **Scott Radke**, Iowa State University; and **Ryan Tenbergen**, University of Guelph.

Student presenters receiving \$500 scholarships were **Colleen Crozier**, North Carolina State University; **Amanda Harris**, Iowa State



Recipient of the \$5000 scholarship for top student presenter during AASV's Student Seminar: Joseph Thomas, Iowa State University. Pictured with Joseph is Shelley Stanford (right) of Zoetis, sponsor of the Student Seminar and top student presenter award.



Kim Lawson (far left) presented scholarships sponsored by Elanco Animal Health. Recipients of the \$2500 AASV Foundation scholarships were (from left) Alyssa Taplett, Iowa State University; Kathleen Wood, North Carolina State University; Brianna Peters, University of Tennessee; Chris Sievers, Iowa State University.



Kim Lawson (far left) presented scholarships sponsored by Elanco Animal Health. Recipients of the \$1500 AASV Foundation scholarships were (from left) Brigitte Mason, University of Illinois; Hunter Baldry, University of Minnesota; Jacqueline Myers, Iowa State University; Ryan Tenbergen, University of Guelph; Scott Radke, Iowa State University.

University; **Alexandra John**, University of Pennsylvania; **Emily Renner**, University of Minnesota; and **Quinn Robinson**, Iowa State University.

Sixty-one veterinary students from 16 universities submitted abstracts for consideration. From those submissions, 15 students were selected to present during the annual meeting. Zoetis, sponsor of the Student

Seminar, provided a \$750 travel stipend to each student selected to participate.

A panel of judges selected the recipients on the basis of communications skills in writing the abstract and the presentation of the case report and on applicability of the research to swine medicine.



Kim Lawson (far left) presented scholarships sponsored by Elanco Animal Health. Recipients of the \$500 AASV Foundation scholarships were (from left) Quinn Robinson, Iowa State University; Colleen Crozier, North Carolina State University; Emily Renner, University of Minnesota; Amanda Harris, Iowa State University (not pictured: Alexandra John, University of Pennsylvania).

AASV announces Veterinary Student Poster Competition awardees

The American Association of Swine Veterinarians (AASV) provided an opportunity for 15 veterinary students to compete for awards in the Veterinary Student Poster Competition at the association's 46th annual meeting in Orlando, Florida. Newport Laboratories sponsored the competition, offering awards totaling \$4000.

On the basis of scores received in the original judging of abstracts submitted for the AASV Student Seminar, the top 15 abstracts not selected for oral presentation at the



Dr Joel Flores (left), representing sponsor Newport Laboratories, congratulates Donna Drebes, University of Minnesota, on winning the top prize of \$500 for best poster.

annual meeting are eligible to compete in the poster competition.

Newport Laboratories announced the following awards during the AASV Luncheon on March 2:

\$500 scholarship: **Donna Drebes**, University of Minnesota, top student poster entitled "The effect of translactational anti-

bodies on preweaning mortality in a porcine epidemic diarrhea virus exposed herd;"

\$400 scholarships: **Tyler Te Grotenhuis**, Iowa State University; **Joseph Yaros**, Cornell University;

\$300 scholarships: Bernadetta Bernatowicz, University of Pennsylvania; Dan Breuer, Iowa State University; Joel Sparks, Iowa State University;

\$200 scholarships: Stephanie Derbawka, University of Saskatchewan; Taylor Engle, Virginia-Maryland Regional College of Veterinary Medicine; Danielle Evenson, Iowa State University; Daniel Gascho, Purdue University; Cassy Griebel, University of Minnesota; Amanda Jara, University of Georgia; Erin Jobman, Kansas State University; Kayla Ohrt, Iowa State University; Chelsea Onken, Iowa State University.

In addition to the poster competition awards, each student poster participant received a \$250 travel stipend from Zoetis and the AASV.

Annual Business Breakfast

American Association of Swine Veterinarians President Dr Michelle Sprague reported on the association's membership and activities during the annual breakfast on Tuesday, March 3. She stated that there were 1741 members, including 329 student members. Dr Sprague thanked outgoing director Dr Bill DuBois (District 7) and Amy



Dr Joel Flores (far left), representing sponsor Newport Laboratories, congratulates the \$400 poster-competition winners (from left) Joseph Yaros, Cornell University, and Tyler Te Grotenhuis, Iowa State University.



Dr Joel Flores (far left), representing sponsor Newport Laboratories, congratulates the \$300 poster-competition winners (from left) Joel Sparks, Iowa State University; Dan Breuer, Iowa State University; and Bernadetta Bernatowicz, University of Pennsylvania.



Dr Joel Flores (far left), representing sponsor Newport Laboratories, congratulates the \$200 poster-competition winners (from left) Kayla Ohrt, Iowa State University; Cassy Griebel, University of Minnesota; Amanda Jara, University of Georgia; Stephanie Derbawka, University of Saskatchewan; Chelsea Onken, Iowa State University; and Daniel Gascho, Purdue University (not pictured: Taylor Engle, Virginia-Maryland Regional College of Veterinary Medicine; Danielle Evenson, Iowa State University; Erin Jobman, Kansas State University).

Daniels, student delegate to the board, for their service. She congratulated incoming Director Dr Scanlon Daniels (District 7) and incoming Alternate Student Delegate Emily Mahan-Riggs. Dr Sprague announced that Dr Lynette Holman was re-elected director for District 1 and that there would be an election to replace Dr Ramirez (District 6), given his election as vice president. Honored guests at the Business Breakfast included Dr Ted Cohn (AVMA President),

Dr Gary Brown (AVMA executive board representative), Dr Paul Sundberg (NPB senior VP of science and technology), and Dr Liz Wagstrom (National Pork Producers council chief veterinarian). The audience heard updates from each respective organization. Approximately 200 people attended the breakfast.

New officers

Dr Ron Brodersen was installed as president, succeeding Dr Michelle Sprague, who is now immediate past president. Dr George Charbonneau has ascended to president-elect. The newly elected vice president is Dr Alejandro "Alex" Ramirez.

Save the date

The 2016 annual meeting is scheduled for February 27 to March 1, 2016, in New Orleans, Louisiana.

Photo courtesy statement

Photos are courtesy of Tina Smith.







Swine veterinarians inform, share, learn



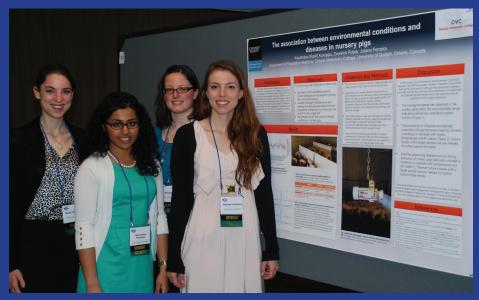


Thank you, AASV Annual Meeting sponsors!

AASV members attending the annual meeting make a substantial investment in the form of registration fees, travel, lodging, meals, and potential loss of income while away from work. However, the cost of attendance would be even greater – or the quality of the meeting experience reduced – if it were not for the financial support provided by corporate sponsors for refreshments, meals, and social activities, as well as scholarships and travel stipends for veterinary students. The AASV extends its sincere appreciation for the sponsorship of meeting events by the following companies:

- Boehringer Ingelheim Vetmedica, Inc (AASV Luncheon)
- CEVA Animal Health (Refreshment Break)
- Elanco Animal Health (AASV Awards Reception and AASV Foundation Veterinary Student Scholarships)
- GlobalVetLINK (Internet Kiosk)
- Harrisvaccines (Refreshment Break)
- Hog Slat (Refreshment Break)
- Merck Animal Health (Student Reception and Student Swine Trivia Event)
- Merial (Refreshment Break)
- Newport Laboratories (Veterinary Student Travel Stipends, Veterinary Student Poster Scholarships, Refreshment Break)
- Stuart Products (Praise Breakfast)
- Zoetis (Welcome Reception, AASV Student Seminar and Student Poster Session, AASV Foundation Top Student Presenter Scholarship)

The AASV is also grateful to the 84 companies and organizations that provided support through their participation in the 2015 Technical Tables exhibit. Thank you all!

















FOUNDATION NEWS

2015 AASV Foundation auction a wild ride

The 2015 AASV Foundation Committee dedicated this year's auction in memory of one of the foundation's strongest supporters, Dr Rod Johnson and his wife Jean. Rod would have been very pleased with everyone's generous donations and inspired bidding that resulted in the foundation raising \$102,585! This marked the third year in a row that support topped \$100,000. This tremendous effort was aided by the generosity of Mary Lou Hogg and MVP Laboratories through the donation of a Harley Davidson motorcycle. The hog raffle raised \$30,150! The funds raised support foundation programs, including student travel stipends, research projects, scholarships, student externships, summer internships, awards, and other opportunities to enhance the personal and professional aspects of swine veterinary medicine.

Auctioneer and AASV Executive Director Dr Tom Burkgren called the auction with the assistance of Dr Shamus Brown and Dr Jess Waddell. The spirited live auction raised \$43,500. This was in addition

to the \$22,185 collected during the silent auction and \$6750 in generous cash donations. The foundation thanks all those who participated in the auction by bidding on or donating items, as well as those who served on the auction committee chaired by Dr Daryl Olsen.

A special thanks goes to the ring men:
Drs Butch Baker, Shamus Brown, Jeff Harker,
Howard Hill, Darrell Neuberger, David
Reeves, Jess Waddell, and John Waddell,
who kept the bids coming. In addition, the
following folks' behind-the-scenes and frontend help were invaluable: Joel Burkgren,
Wes Johnson, Kay Kimpston-Burkgren, Sue
Kimpston, Karen Menz, Karen Richardson,
Lee Schulteis, Sue Schulteis, Tina Smith, and
Harry Snelson.

An extra-special thanks goes out to Lee Schulteis for driving the truck and trailer, containing all the auction items and meeting materials, from Perry, Iowa, to Orlando and back again.



After drawing the winning raffle ticket for the Harley Davidson motorcycle, Mary Lou Hogg, MVP Laboratories, passes the keys to the raffle winner, Gentrie Shafer

Photo courtesy of Tina Smith

The AASV Foundation Auction Committee is grateful to everyone who purchased a raffle ticket or bid on items in the live and silent auctions. We are pleased to recognize and thank the following bidders who purchased one or more items at the auction:

Matt Anderson
Paul Armbrecht
John Baker
Joel Burkgren
Larry Coleman
Dennis Dwyer
Mark Engesser
Bob Evelsizer
Corky Feuerbach
Glenn Gaines
Doug Groth
Peggy Anne Hawkins
Dale Hendrickson
Daniel Hendrickson
Bill Hollis

Micah Jansen
Jean Johnson
Randy Jones
Kerry Keffaber
Barry Kerkaert
Paul Knoernschild
Jim Kober
Seth Krantz
Beth Lautner
Ian Levis
Duane Long
Ruth Loula
Tim Loula
Erin Lowe
Tom Marsteller

Dale Mechler
Michelle Michalak
Bill Minton
Theresa Minton
Jana Morgan
Betsy Newton
Daryl Olsen
Tom Petznick
Mike Pierdon
Doug Powers
Jessica Rosener
Paul Runnels
Brian Schantz
Kent Schwartz

Mike Senn Cliff Smith Gordon Spronk Deb Sundberg Paul Sundberg Mike Terrill Pete Thomas Lisa Tokach Dennis Villani Douglas Weiss Teddi Wolff Kathleen Wood

AASV Foundation news continued on page 169





Brief Summary

CAUTION: Federal (USA) law restricts this drug to use by or on the order of a licensed

DESCRIPTION

BESCRIPTION

CHARACTH 25 Injectable Solution is a ready-to-use sterile parenteral preparation containing utathromycin, a semi-synthetic macrolide antibiotic of the subclass triaming to the subclass triaming the subclass triaming the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming the subclass triaming to the subclass triaming the subclass triaming the subclass triaming to the subclass triaming the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming to the subclass triaming triaming the subclass triaming the subclass triaming to the subclass triaming triaming the

INDICATIONS

Minute State

DRAXXII 25 Injectable Solution is indicated for the treatment of swine respiratory dissees (SRD) associated with Actinobacillus pleuropneumoniae, Pasteurella multiocida,
Bordetella bronchiseptica, Haemophilus parasus, and Mycoplasma hyopneumoniae,
and for the control of SRD associated with Actinobacillus pleuropneumoniae, Pasteunultocida, and Mycoplasma hyopne niae in groups of pigs where SRD has

Suckling Calves, Dairy Calves, and Veal Calves BBD - DRAXXIN 25 Injectable Solution is indicated for the treatment of bovine respiratory disease (BRD) associated with Mannheima haemolytica, Pasteurella multo-cida, Histophilus somni, and Mycoplasma bovis.

DOSAGE AND ADMINISTRATION

Swine Inject intramuscularly as a single dose in the neck at a dosage of 2.5 mg/kg (1 mL/22 lb) Body Weight (BW). Do not inject more than 4 mL per injection site.

Table 1. DRAXXIN 25 Swine Dosing Guide (25 mg/mL)

	J (. J)
Animal Weight (Pounds)	Dose Volume (mL)
4	0.2
10	0.5
15	0.7
20	0.9
22	1.0
25	1.1
30	1.4
50	2.3
70	3.2
90	4.0

Carrest inject subcutaneously as a single dose in the neck at a dosage of 2.5 mg/kg (1 mL/22 lb) body weight (BW). Do not inject more than 11.5 mL per injection site.

Involve 20 dail booking dalac (20 mg/m2)	
Animal Weight	Dose Volume
(Pounds)	(mL)
50	2.3
75	3.4
100	4.5
150	7.0
200	9.0
250	11.5

CONTRAINDICATIONS

The use of DRAXXIN 25 Injectable Solution is contraindicated in animals previously found to be hypersensitive to the drug.

WARNINGS FOR USE IN ANIMALS ONLY. NOT FOR HUMAN USE. KEEP OUT OF REACH OF CHILDREN. NOT FOR USE IN CHICKENS OR TURKEYS.

RESIDUE WARNINGS Swine Swine Swine intended for human consumption must not be slaughtered within 5 days from the last treatment.

lves
ves intended for human consumption must not I
days from the last treatment with DRAXXIN 25 Inj drug is not for use in ruminating cattle

PRECAUTIONS

Swine Cs of Draxin 25 Injectable Solution on porcine reproductive performance, The effects of Draxin 25 Injectable Solution on porcine reproductive performance, pregnancy, and lactation have not been determined. Intramuscular injection can cause a transient local tissue reaction that may result in trim loss of edible tissue at slaughter.

a transient local tissue reaction that may result in trim loss of edible tissue at slaughter. **Cattle**The effects of Draxxin 25 Injectable Solution on bovine reproductive performance, pregnancy, and lactation have not been determined. Subcutaneous injection can cause a transient local tissue reaction that may result in trim loss of edible tissue at slaughter

ADVERSE REACTIONS

Swine In one field study, one out of 40 pigs treated with DRAXXIN Injectable Solution (100 mg/ mL) at 2.5 mg/kg BW exhibited mild salivation that resolved in less than four hours. Calves In one BRD field study, two calves treated with DRAXXIN Injectable Solution (100 mg/ mL) at 2.5 mg/kg BW exhibited transient hypersalivation. One of these calves also exhibited transient dyspnea, which may have been related to pneumonia. Post Approval Experience

Post Approval Experience
The following adverse events are based on post approval adverse drug experience
reporting for DRAXXIN injectable Solution (100 mg/mL). Not all adverse events reported to the FDA CVML it is not always possible to reliably estimate the
adverse event frequency or establish a causal relationship to product exposure using
these data. The following adverse events are listed in decreasing order of reporting
frequency in cattle: Injection site reactions and anaphylaxis/anaphylaxitoir reactions. For
a complete Isting of adverser aexistors for PRAXXIN injectable Solution or DRAXXIN 25 Injectable Solution reported to the CVM see: http://www.fda.gov/AnimalVet NADA 141-349, Approved by FDA

zoetis

Distributed by: Zoetis Inc. Kalamazoo, MI 49007

To report a suspected adverse reaction or to request a safety data sheet call 1-888-963-9471. For additional information about adverse drug experience reporting for animal drugs, contact PDA at 1-888-PBA-VETS or online at http://www.flaga-valimal/veterina/setely/health.

For additional DRACKIN 25 product information calls: 1-888-DRAXXIN or go to www.DRAXXIII.00016.



Made in Brazil

060005AAA&P Revised: September 2014

Hogg Scholarship

Dr Chris Rademacher was named the 2015 recipient of the American Association of Swine Veterinarians Foundation Hogg Scholarship. The scholarship was presented by Mary Lou Hogg during the association's annual meeting in Orlando, Florida.

Established in 2008, the scholarship is named for Dr Alex Hogg, who was a leader in swine medicine and pursued a master's degree in veterinary pathology after 20 years in a mixed-animal practice. The scholarship is awarded annually to an AASV member who has been accepted into a qualified graduate program to further his or her education after years as a swine practitioner.

Dr Rademacher earned his DVM from the University of Minnesota in 1998. Following graduation, he joined New Fashion Pork as the staff veterinarian and service manager. He became the Director of Health Services and spent 10 years with the company before joining Murphy-Brown Western Operations as Director of Production Improvement. In December, 2014 he accepted the position of Swine Extension Veterinarian at Iowa State University (ISU). He recently enrolled in the Master's in Veterinary Preventive Medicine program at ISU under Dr Jeff Zimmerman.

Rademacher recently received a grant to study porcine epidemic diarrhea virus (PEDV) vaccination and immunity. The objective of the study is to evaluate the production benefit of vaccination in an endemically infected sow herd. In addition, the project will attempt to correlate PEDV immunity with production impacts including pre-wean mortality and weaning weight.

When asked about the significance of the scholarship, Dr Rademacher said, "It is indeed an honor and a humbling privilege to be awarded this scholarship that bears the name of one of the legends of swine production medicine. Alex is an inspiration to many of us who want to give back to an industry that has so richly blessed us, and this award allows me to continue my lifelong learning as Alex so wonderfully modeled in his lifetime. I am so thankful to the Hogg family for sponsoring this award annually to swine veterinarians who want to continue their education."



Mary Lou Hogg presents the Alex Hogg Scholarship award to Dr Chris Rademacher during the AASV Annual Meeting

Photo courtesy of Tina Smith

AASV Foundation announces research funding for 2015

The AASV Foundation selected four research proposals to receive a total of \$60,000 in funding for 2015. The research will study a wide range of topics important to swine veterinarians, including education, welfare, porcine epidemic diarrhea virus (PEDV), porcine reproductive and respiratory syndrome virus (PRRSV), and porcine circovirus (PCV). Dr Daryl Olsen, chairman of the AASV Foundation, announced the proposals selected for funding during the foundation's annual luncheon on March 1 in Orlando, Florida.

A grant of \$7490 was awarded to Dr Paisley Canning at Iowa State University (ISU) to fund a proposal designed to help address the challenge of providing swine training to interested swine-focused students at veterinary colleges across the country. Dr Canning will develop a network of swine and production-animal medicine clubs at veterinary schools for the purpose of transmitting live broadcasts of three swine-focused seminars hosted by the ISU AASV Student Chapter to the participating schools.

Dr Trevor Schwartz at Suidae Health & Production was awarded a grant of \$16,450 to assist with funding for a study on the effect of PEDV vaccination on PEDV-naive sows and previously PEDV-exposed sows in a controlled PEDV challenge model. The study will compare vaccinated and unvaccinated sow immune response and litter pre-weaning morbidity and mortality in an effort to determine if there is a significant difference between naive and previously exposed sows, and whether the measured immune response can be correlated with litter pre-weaning morbidity and mortality.

The foundation allocated \$15,000 to fund a proposal submitted by Dr Carissa Odland at Pipestone Veterinary Clinic to perform



Research grant recipients (left to right) Drs Paulraj Lawrence, Jennifer Stevens (accepting for Carissa Odland), Paisley Canning, and Trevor Schwartz with Dr Daryl Olsen, chairman of the AASV Foundation

Photo courtesy of Tina Smith

efficacy testing of a novel method of euthanasia of suckling piglets. The method utilizes an aerosol canister to administer isoflurane, then carbon monoxide, into a containment chamber to achieve humane euthanasia. The ultimate goal is to gain Food and Drug Administration approval of the process in order to provide swine veterinarians and producers with a simple and humane method of euthanizing small piglets.

The fourth research grant was awarded to Eric Bumgardner and Dr Paulraj Lawrence at Newport Laboratories. The investigators were awarded \$21,060 to assist with the development of a bivalent PRRSV-PCV type 2 vaccine capable of inducing broader cross-protection against PRRSV. They hope to develop vaccination strategies that can be used in commercial swine herds to protect

animals against diverse PRRSV strains and to gain insight into the immunological properties of PRRSV.

Drs Peggy Anne Hawkins and Nathan Winkelman co-chaired the scientific subcommittee responsible for reviewing and scoring the proposals received for consideration, and they join the foundation in thanking Drs John Baker, Jeff Blythe, Jane Christopher-Hennings, Cate Dewey, and Tom Gillespie for their service on the subcommittee.

An overview of past and current projects funded by the foundation is available at https://www.aasv.org/foundation/research.htm. The foundation will issue its next call for research proposals in the fall of 2015.



Emerging disease, emerging solutions

As evidenced by the 2013 outbreak of porcine epidemic diarrhea virus (PEDV), emerging diseases can have a dramatic effect on swine health and pork production. "Emerging" disease is a broad term for the appearance of pathogens or syndromes not previously known to be causing disease in the national swine herd or the recognition of a significant change in clinical presentation of an endemic pathogen. Either of these scenarios has the potential to impact animal health and well-being, trade, production parameters, food safety, and human health.

Our ability to rapidly recognize and implement an appropriate response can have a significant impact on the outcome of a disease outbreak. The effectiveness of our ability to recognize and respond is largely dependent on our preparation, including acknowledging the existing threats and having a mechanism in place to identify and address known resource gaps. In order to be prepared for future emerging diseases, we need to identify potential sources of information; capture, coordinate, and research that information; and, finally, act on the information. Let's first discuss the numerous sources of information available to us.

A number of years ago, the American Association of Swine Veterinarians (AASV) and

the National Pork Board (NPB) established the Sentinel Veterinary Clinic program. In an effort facilitated by the NPB and Iowa State University's Center for Food Safety and Public Health, a group of select veterinary clinics meets by phone with representatives of the key diagnostic laboratories on a quarterly basis to discuss clinical observations and diagnostic submissions of interest. This was the first coordinated effort to establish a systematic approach to recognize emerging disease syndromes within the national swine herd. This program arose out of an outbreak of erysipelas that spread through the Midwest, resulting in significant increases in morbidity and mortality. Samples were being submitted to multiple laboratories by multiple practitioners, but we were slow to realize the significance and distribution of the disease. This outbreak might have been averted if a system had been in

"Anecdotal reports often provide a real-world glimpse into the diseases circulating in domestic and international herds. These reports need to be captured and investigated."

place to facilitate communication between practitioners and diagnostic laboratories.

Although veterinarians, producers, and the diagnostic laboratories responded quickly to the PEDV diagnosis in May 2013, discussion about PEDV circulation in China had been going on in various forums for quite some time prior to the virus actually entering the United States. Unfortunately, there was no formal system in place to gather that intelligence and

do anything about it. The AASV, NPB, and the National Pork Producers Council (NPPC) are taking steps to change that.

In 2014, the AASV Swine
Health Committee was charged
with evaluating and prioritizing
a list of all known swine viruses
and identifying resources needed
to diagnose and respond to each
disease if it should be introduced

into the North American swine herd. The first phase of this on-going process has been completed, resulting in development of a swine virus matrix. This matrix provides an initial prioritization of the viruses and outlines the key resource categories that need to be addressed to facilitate response planning.

The AASV and pork producers are collaborating with the Institute for Infectious Animal Diseases at Texas A&M University to develop a Web-based syndromic disease surveillance application for practitioners to use in the field. Termed Enhanced Passive Surveillance (EPS), this project will ultimately provide a coordinated electronic system by which practitioners' data can be securely and confidentially combined with other observations and laboratory data to facilitate early recognition of emerging syndromes of consequence to the US swine herd. An EPS pilot project should be underway by the time you read this article.

In addition, numerous AASV members, pork producers, researchers, allied industries, and government officials have formal and informal contacts providing insight into global swine-disease challenges. Researchers and diagnosticians around the world frequently publish informed reports on diseases impacting swine herds in their countries. Anecdotal reports often provide a real-world glimpse into the diseases circulating in domestic and international herds. These reports need to be captured and investigated.

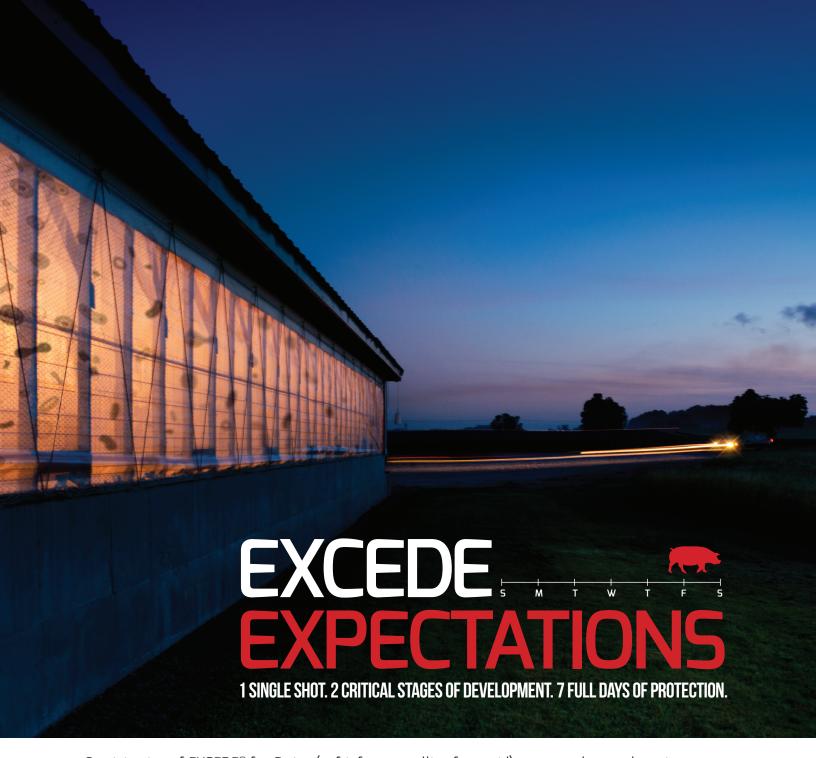
All of this provides a potential wealth of information if we can find a way to coordinate it and channel it to the right people. That's where the Swine Health Information Center (SHIC) comes in. The SHIC has been under discussion for about a year and is now coming to fruition. The NPB Board of Directors recently committed to a one-time investment of \$15 million spread over the next 5 years to fund the SHIC.

The center's objectives are threefold. First, the center will monitor foreign and endemic

e center will monitor foreign and endemic



Advocacy continued on page 173



One injection of EXCEDE® for Swine (ceftiofur crystalline free acid) treats and controls swine respiratory disease for 7 days. It continuously attacks a broad range of pathogens.* And EXCEDE is proven effective for both weaning and nursery—the 2 critical stages in a young pig's development. So you can have a healthy pig—and a healthy herd—for the long term.

IMPORTANT SAFETY INFORMATION: People with known hypersensitivity to penicillin or cephalosporins should avoid exposure to EXCEDE. Do not use in swine found to be hypersensitive to the product. Pre-slaughter withdrawal time is 14 days following the last dose. See Brief Summary of Prescribing Information on the next page.

*A pleuropneumoniae, H parasuis, P multocida, S suis.







For intramuscular administration in the post-auricular region of the neck of swine.

CAUTION

Federal (USA) law restricts this drug to use by or on the order of a licensed veterinarian.

INDICATIONS

EXCEDE FOR SWINE Sterile Suspension 100 mg/mL is indicated for the treatment of swine respiratory disease (SRD) associated with Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis, and Streptococcus suis; and for the control of SRD associated with Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis, and Streptococcus suis in groups of pigs where SRD has been diagnosed.

CONTRAINDICATIONS

As with all drugs, the use of EXCEDE FOR SWINE Sterile Suspension 100 mg/mL is contraindicated in animals previously found to be hypersensitive to the drug.

WARNINGS

FOR USE IN ANIMALS ONLY. Not for human USE. Keep out of Reach of Children.

Penicillins and cephalosporins can cause allergic reactions in sensitized individuals. Topical exposures to such antimicrobials, including ceftiofur, may elicit mild to severe allergic reactions in some individuals. Repeated or prolonged exposure may lead to sensitization. Avoid direct contact of the product with the skin, eyes, mouth and clothing. Sensitization of the skin may be avoided by wearing protective gloves.

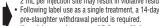
Persons with a known hypersensitivity to penicillin or cephalosporins should avoid exposure to this product.

In case of accidental eye exposure, flush with water for 15 minutes. In case of accidental skin exposure, wash with soap and water. Remove contaminated clothing. If allergic reaction occurs (e.g., skin rash, hives, difficult breathing), seek medical attention.

The material safety data sheet contains more detailed occupational safety information. To report adverse effects in users, to obtain more information or to obtain a material safety data sheet, call 1-800-366-5288.

RESIDUE WARNINGS

 A maximum of 2 mL of formulation should be injected at each injection site. Injection volumes in excess of 2 mL per injection site may result in violative residues.



pre-slaughter withdrawal period is required.

• Use of dosages in excess of 5.0 mg ceftiofur equivalents (CE)/kg or administration by an unapproved route may result in illegal residues in edible tissues.

PRECAUTIONS

The safety of ceftiofur has not been demonstrated for pregnant swine or swine intended for breeding.

Administration of EXCEDE FOR SWINE Sterile Suspension 100 mg/mL as directed may induce a transient reaction at the site of injection and underlying tissues that may result in trim loss of edible tissue at slaughter.

ADVERSE REACTIONS

An injection site tolerance study demonstrated that EXCEDE FOR SWINE Sterile Suspension 100 mg/mL is well tolerated in pigs. Half of the injection sites at both 3 and 7 days post-injection were scored as "negative" for irritation and the other half were scored as "slight irritation". All gross observations and measurements of injection sites qualified the sites at 10 days post-injection as "negative" for irritation.

No adverse effects were observed in multi-location field efficacy studies involving more than 1000 pigs.

STORAGE CONDITIONS

Store at controlled room temperature 20° to 25°C (68° to 77°F). Shake well before using. Contents should be used within 12 weeks after the first dose is removed.

HOW SUPPLIED

EXCEDE FOR SWINE Sterile Suspension 100 mg/mL is available in the following package size:

100 mL vial

NADA #141-235, Approved by FDA



Pharmacia & Upjohn Company Division of Pfizer Inc, NY, NY 10017

www.PFIZERPORK.com or call 1-866-387-2287

Revised: March 2010

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disease risks and vulnerabilities by collecting swine-disease risk information from, among others, the sources outlined above. This information will help inform producers and veterinarians about emerging swine-disease risks. Second, the center will fund and manage research to fill knowledge gaps identified in the swine virus matrix. This will focus resources in a prioritized manner to provide the tools necessary to diagnose and respond to emerging diseases. Third, the center will support epidemiological analysis of emerging swine diseases and coordination of domestic swine herd-health information to support international trade of US pork products.

The SHIC leadership is a collaborative effort involving AASV, NPB, and NPPC. Each organization has two representatives on the SHIC Board of Directors. Drs Matt Anderson and Daryl Olsen represent AASV. In addition to those six members, there are three at-large producer members (Mark Schwartz, Sleepy Eye, Minnesota; Mike Terrill, Burnsville, Minnesota; and Matthew Turner, Clinton, North Carolina). The objectives of the center are designed to complement, and not duplicate, the efforts and responsibilities of the three organizations.

Now that we have collected all this information and directed resources to identify and address gaps in our knowledge base, how do we act? The decision to respond or not is again a collaborative one. It involves state, federal, and industry coordination. The NPPC is working to stand up a collaborative board similar in design to the Pseudorabies Virus (PRV) Control Board that functioned to provide input on program standards during the PRV eradication effort. The idea would be that this board would evaluate emerging disease issues and strive to offer response guidance on the basis of a consensus of impacted stakeholder groups.

All of these efforts will serve to increase our readiness for the next emerging disease that threatens the swine industry. Preparedness is the key to success. As the author Stephen King once said, "there's no harm in hoping for the best as long as you're prepared for the worst.\(^{1}\)"

Reference

1. King, S. *Different Seasons*. New York, New York: Signet (division of Penguin Group); 1982.

Harry Snelson, DVM Director of Communications



2015 AASV Foundation Golf Outing

Thursday, August 20, 2015 11:00 AM – 6:00 PM











Landsmeer Golf Club

902 7th Street NE • Orange City, IA 51041 www.landsmeergolfclub.com

https://www.aasv.org/foundation

UPCOMING MEETINGS

World Pork Expo

June 3-5, 2015 (Wed-Fri)

Iowa State Fairgrounds, Des Moines, Iowa

For more information:

Alicia Newman

National Pork Producers Council

10676 Justin Drive, Urbandale, IA 50322

Tel: 515-864-7989; Fax: 515-278-8014

E-mail: irlbecka@nppc.org
Web: http://www.worldpork.org

International PRRS Congress

June 3-5, 2015 (Wed-Fri)

Ghent, Belgium

For more information: E-mail: prrs2015@ugent.be

Web: http://www.prrscongress.ugent.be/

7th International Symposium on Emerging and Re-emerging Pig Diseases

June 21-24, 2015 (Sun-Wed)

Kyoto International Conference Center, Kyoto, Japan

For more information:

E-mail: iserpd20150ics-inc.co.jp Web: http://emerging2015.com

VIIIth International Conference on Boar Semen Preservation

August 9-12, 2015 (Sun-Wed)

Hilton Garden Inn, Urbana-Champaign, Illinois

For more information:

Web: http://boarsemen2015.com/

Passion for Pigs "Learn to Earn" Tour

August 25, 2015 (Tue): Cedar Rapids, Iowa September 2, 2015 (Wed): St Louis, Missouri November 3, 2015 (Tue): Dayton, Ohio November 19, 2015 (Thu): Orange City, Iowa December 8, 2015 (Tue): Columbia, Missouri

For more information:

Julie A. Lolli, Executive Coordinator

Tel: 660-657-0570

E-mail: julie.nevets@nevetsrv.com Web: http://www.passionforpigs.com

2015 Allen D. Leman Swine Conference

September 19-22, 2015 (Sat-Tue) St Paul RiverCentre, St Paul, Minnesota

For more information:

University of Minnesota

Veterinary Continuing Education

1365 Gortner Avenue, St Paul, MN 55108

 $Web: { t http://www.cvm.umn.edu/vetmedce/events/adl/home.html}$

5th International Symposium on Animal Mortality Management

September 28-October 1, 2015 (Mon-Thu)

Lancaster Marriott at Penn Square, Lancaster, Pennsylvania

For more information:

Heather Simmons

Institute for Infectious Animal Diseases

Tel: 979-845-2855

E-mail: hsimmons@ag.tamu.edu

Dale Rozeboom

Michigan State University

Tel: 517-355-8398

E-mail: rozeboom@msu.edu

Web: http://animalmortmgmt.org

The 4th Leman China Swine Conference

October 11-13, 2015 (Sun-Tue)

Nanjing, China

Program Director: Frank Liu Veterinary Diagnostic Laboratory,

1333 Gortner Avenue, St Paul, MN 55108

Tel: 612-625-2267 Fax: 612-624-8707

E-mail: liuxx063@umn.edu

Web: http://www.cvm.umn.edu/lemanchina/

American Association of Swine Veterinarians $47^{\rm th}$ Annual Meeting

February 27-March 1, 2016 (Sat-Tue)

Hyatt Regency New Orleans, New Orleans, Louisiana

For more information:

American Association of Swine Veterinarians

830 26th Street, Perry, IA 50220-2328

Tel: 515-465-5255 Fax: 515-465-3832 E-mail: aasv@aasv.org

Web: http://www.aasv.org/annmtg

24th International Pig Veterinary Society Congress

June 6-10, 2016 (Mon-Fri)

Dublin, Ireland

For more information:

Web: http://www.ipvs2016.com





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Colorful Iberian commanche sow

Photo courtesy of Dr Antonio Palomo Yague

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