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AASV
830 26th Street, Perry, IA 50220-2328
Tel: 515-465-5255; Fax: 515-465-3832
E-mail: aasv@aasv.org

Editorial questions, comments, and inquiries should be addressed to Karen Richardson, Publications Manager:
Tel: 519-856-2089; Fax: 519-763-3117;
E-mail: pub_mgr@aasv.org

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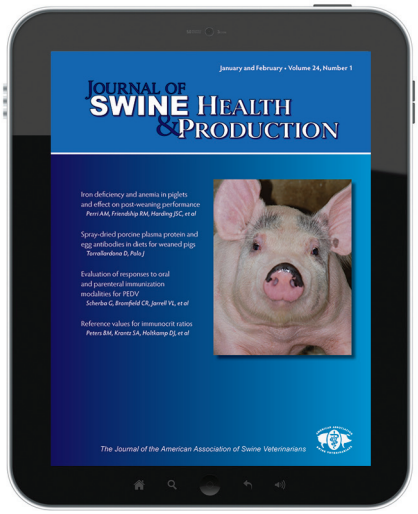

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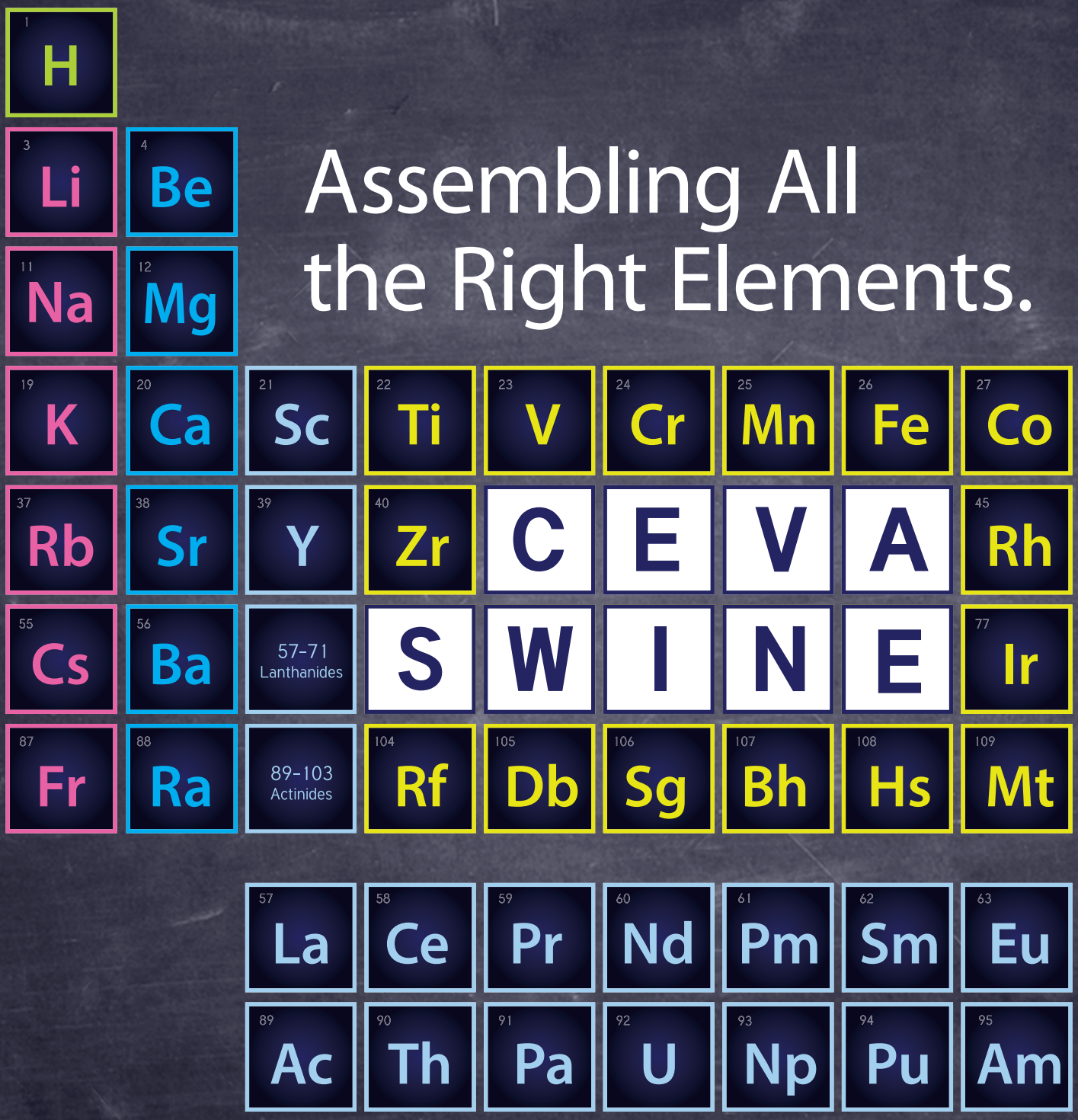
quoted from President's message, page 5



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The irony of biosecurity – time to open the curtain

Pig veterinarians understand biosecurity. Pig producers understand biosecurity. "Town people" not so much.

When I drive onto a client's farm, I always slip on disposable shoe covers as I step out of my vehicle. This is followed by a change into the boots and clothes provided by the farm. When I am back in town, I also slip on disposable shoe covers at the gas station, farm store, or other locations frequented by farmers. Returning to my vehicle I pull off the shoe covers, turn them inside out and place them in a garbage bag behind the seat so my floor mat doesn't get contaminated. People in town look at me like I have some kind of "obsessive-compulsive-disorder (OCD)." I just smile and say "I am preventing spread of that new pig virus with my shoe condoms." Some laugh, and some look even more puzzled. If they saw my office desk they would know I do not have OCD!

Farm biosecurity is a series of management practices designed to minimize or prevent the introduction of infectious diseases onto a farm. Our non-farm friends do not understand the concept of farm biosecurity. (My spell checker highlights "biosecurity" as a misspelled word). From their own human-health experiences they understand prevention of disease by avoiding direct contact. But they do not understand the concept of indirect transmission through contamination and movement of inanimate objects. The concept is easy to learn, and when you explain it, they get it. They can relate to "dirty boots," "door knobs," and "TV remote controls."

At the farm, good biosecurity management practices include farm entry protocols limiting "drive-in" and "walk-in" visitors. Farms have fences and big signs that say "no visitors allowed." Veterinarians have been so effective in teaching pig farmers the principles of good biosecurity that they have created a "curtain," limiting farm access to some of the greatest allies of animal agriculture – our cousins, neighbors, and friends.

> *"We can help our clients rebuild trust by assisting the industry in developing biosecure means and methods to allow visitors."*

Ironically, we have helped the adversaries of animal agriculture by holding up the curtain that they site as evidence for distrust, when in reality, the curtain is there not to hide transgressions, but to prevent disease. Really, we know that there are not that many anti-meat extremists. Yet they have successfully recruited sympathizers on welfare and antibiotic issues for which we know there are good solutions. Sympathizers recruited have money and social media presence – and we have played right into it. Some sympathizers are American Veterinary Medical Association (AVMA) members!

Our AASV association and its members are objective, trusted, and well respected. We share positions of authority inside the AVMA on many issues, including welfare and antibiotic issues. We are also problem solvers. I believe it is time AASV and its members solve the problem of the biosecurity curtain. We can help our clients rebuild trust by assisting the industry in developing biosecure means and methods to allow visitors. I see it already happening. Some farms are hosting an annual "open house" for their community. Fair Oaks Farms of Indiana's Pig Adventure has been a huge public-relations success. Last year I was a guest of DNA Swine Genetics in Nebraska at their Insight Performance Center where I was privileged to see the investment they had made into their Observatory Conference Room viewing the inside of their production facility.

All of these are great ideas! We can do more. Operation Main Street, sponsored by the National Pork Board, has attracted many of our members to participate in public outreach to "get the word out" with public-speaking opportunities. Just ask Dr Jeff Harker. What a great promoter he is! We need more "Jeffs"! Wouldn't it be great if we could get Operation Main Street in front of every veterinary student in the country?

Better yet would be for every veterinary student to be on a hog farm at least once. That would be a concrete measurable achievement. We need future AVMA members to understand the swine industry and what pig veterinarians do. Let's make the biosecurity curtain transparent.

Ron Brodersen, DVM
AASV President

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New Year's resolutions

Happy New Year JSHAP readers! I really have never been one to make a New Year's resolution. Perhaps it is a subconscious defence mechanism, as I anticipate I would be in the population of people who don't meet their resolutions. But, as a busy academic and veterinarian, I have been thinking about what a reasonable resolution for me should or could be for general self-improvement. In today's fast-paced society of activities with high time demands, deadlines, work demands, and social pressures, I wonder how all of you establish a healthy work-life balance. Do you make a New Year's resolution? Are you (or people in general) willing to share resolution stories?

This tweaked my statistical curiosity to see if there was any information available about how many people make a resolution, who sticks to it, and what those resolutions might be. Not surprisingly, the Internet came through for me and I found some numbers – my disclaimer here is that these statistics are likely full of bias, confounding, and other statistical no-no's, but I thought it would still be fun to look at them. Some of the top 10 resolutions, as listed by Statistic Brain Research Institute,[1] include the following: tame the bulge (lose weight), tame the clutter (get organized), feed the brain (learn something new), spend more time with family, and help others. I couldn't find a list that specifically stated work less or maintain a healthy work-life balance – perhaps spending more time with family is a proxy for work less?

"In today's fast-paced society of activities with high time demands, deadlines, work demands, and social pressures, I wonder how all of you establish a healthy work-life balance."

Then the statistics get more interesting. The same report states that almost half of Americans usually make a New Year's resolution, with 38% reporting that they never or infrequently make a resolution. Only 8% of people reported being successful at achieving their resolution, with repeat offenders (people who repeatedly fail at achieving their resolution) at 24%. Apparently 39% of people in their twenties are more likely to achieve their resolution each year, versus the 14% of successful resolution keepers in their fifties. Even though I have been saying "I am 29 and holding" for many years now, I am not in the age category with high success rate for achieving a resolution. So how can I be successful at keeping a resolution if I make one?

I just finished a series of lectures with the first-year DVM students. When I teach the health management cycle in our food-producing animal lectures, I talk about helping producers make SMART goals: Specific, Measureable, Attainable, Realistic, and Timely. I think most of you would agree that this is a successful strategy when counselling a client through a farm-health or production problem. I thought that this model could be applied to resolution-making and resolution attainability and perhaps then to general self-improvement. I am actually writing this editorial in November 2015 (trying to meet our publisher's deadline), so I am going to think further about making a New Year's resolution and applying a SMART goal (or goals) to increase the likelihood of my success. Perhaps making a New Year's resolution is not for you. But I am going to give it serious consideration this year.

All the best to all of you for 2016!

Reference

1. Statistics Brain Research Institute. Available at **http://www.statisticbrain.com/new-years-resolution-statistics/**. Accessed 18 November 2015.

Terri O'Sullivan, DVM, PhD
Executive Editor

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Why I do what I do

I was born in Mexico City, but from a young age I liked open spaces. When I was 8 years old, my family moved to the south of the city, where it was less populated. Around my home were corn fields, with cattle and sheep grazing openly everywhere, owned mainly by small producers who milked 10 to 50 cows. On Saturdays, an open market sold farm animals, from donkeys to cows, sheep, goats, pigs, and hens.

After finishing college, I applied to the veterinary school at the National University of Mexico. At that time, the Food and Agriculture Organization (FAO) had a postgraduate program, and many of my professors were veterinarians returning with masters or PhD degrees from universities all around the world, so I got a first-quality veterinary education. As a student, I was invited by Dr Aline Shunemann to join the pathology department, where she was head. Under her supervision and example, I not only learned pathology, but developed an interest in understanding why animals become sick and die, and a willingness to accept challenges. Nothing is true unless you can prove it. Teaching young veterinarians has been my passion.

Originally, I wanted to be a large-animal veterinarian working with cattle, but eventually I found swine medicine more challenging, and it became my specialty. When I finished my veterinary degree (DVM), I received a scholarship to study pathology at the Royal Veterinary College in England. I graduated with a master's degree, studying central nervous system (CNS) lesions in experimental cytomegalovirus infection in piglets, comparing the lesions with those caused by other swine viruses. This experience later helped me to identify a new CNS swine pathogen, "blue eye paramyxovirus." You need to be prepared to recognize and understand something new and different.

"Sometimes I would wake up at night with the answer to my questions or with the diagnosis I was looking for."

When I returned to Mexico, the ministry of agriculture was opening regional diagnostic laboratories to form a network covering different regions of the country, and I applied for a position as pathologist at the central diagnostics laboratory. Research was my life. I wanted to understand how infectious and non-infectious agents act to produce disease. Sometimes I would wake up at night with the answer to my questions or with the diagnosis I was looking for.

Since I graduated, many new pathogens have appeared, and some that had been considered opportunists have become recognized as important pathogens. I learned how production systems modified the behavior of some pathogens, and how swine veterinary professionals had moved from clinical practice to swine production. We needed to learn new skills and understand the importance of production practice and the environment on disease control and prevention. I have had many challenges, but the most important one has been porcine reproductive and respiratory syndrome (PRRS) virus: it always has the last word. Just when you think that you know how to control it, something new happens. While it is surprising how much information we achieved in a short time to understand this disease, some questions still need to be answered to control and eradicate PRRS. In my opinion, a good vaccine (heterologous, potent, innocuous, and effective) and a test that allows differentiation of antibodies from natural infection and antibodies of vaccination are still pending.

Over time, Cristina, my best partner in life, married me and we raised four great kids. We wanted to give them the opportunity to follow their dreams, so I had to leave my research at the university to get the resources to send them to university. The most difficult decision in my life was leaving my research and my students at the university, but family is first.

I became an international consultant on disease control and swine production, which gave me the opportunity to visit most of the world's swine production areas and to enjoy meeting new people and making new friends. By comparing production systems in different environments and seeing the work of other professionals, my learning experience was continuing.

I have had the opportunity to participate in professional organizations, including the Mexican Swine Veterinary Association, the International Pig Veterinary Society (where I am past president), and the AASV (where I recently served as District 10 representative). All this has given me a sense of pride in being part of a great profession. Taking part in the AASV has helped me to grow and learn. It is an excellent, well-organized swine veterinary association, by far the largest in the world. The AASV participates in swine production and disease control, working alongside researchers, producers, and government agencies, keeping lines of communication open with the public and helping to make the best use of information in any problem or situation that arises. The association's constant support of research and efforts to encourage a new generation of veterinarians will guarantee the long life of this association.

Alberto Stephano, DVM, MSc
International Swine Consultant

ORIGINAL RESEARCH PEER REVIEWED

An investigation of iron deficiency and anemia in piglets and the effect of iron status at weaning on post-weaning performance

Amanda M. Perri, BSc, MSc; Robert M. Friendship, DVM, MSc, Diplomate ABVP; John C. S. Harding, DVM, MSc, Diplomate ABVP; Terri L. O'Sullivan, DVM, PhD

Summary

Objectives: To determine iron status of pigs at weaning and its effects on post-weaning performance, and to determine whether high concentrations of zinc oxide (ZnO) in feed are associated with postweaning anemia.

Materials and methods: A small, medium, and large piglet (N = 1095) were selected per litter 1 to 2 days before weaning from 20 Ontario (Canada) swine farms. Serum and whole blood samples and body weights were collected. Three weeks later, a second body weight and blood sample were collected from the same pigs. Hemoglobin (Hb) and other blood parameters were analyzed to assess iron status and associations with post-weaning performance. Iron supplementation protocols and ZnO concentrations in nursery feed were collected.

Results: Anemic and iron-deficient pigs presented at weaning on most participating farms. Pigs that had been anemic at weaning were 0.82 kg lighter 3 weeks post weaning than piglets that had normal Hb values at weaning ($P < .05$). Larger piglets at weaning had lower red cell parameters and serum iron, and higher total iron binding capacity, than smaller piglets (all $P < .05$). More pigs were anemic 3 weeks post weaning than at weaning ($P < .05$), and prevalence of anemia was associated with high ZnO concentrations ($P < .05$).

Implications: Iron supplementation protocols used in the study herds were inadequate to prevent iron deficiency, particularly in the largest pigs. Anemic pigs at weaning have slower growth rates in the nursery. Consumption of nursery starter feeds containing high concentrations of ZnO is associated with post-weaning anemia.

Keywords: swine, anemia, iron, nursery performance, hemoglobin

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Resumen - Una investigación de la deficiencia de hierro y anemia en lechones y el efecto del estado de hierro en el desempeño post destete

Objetivos: Determinar el estatus de hierro en cerdos al destete y sus efectos en el desempeño post destete, y determinar si las altas concentraciones de óxido de zinc (ZnO) en el alimento están relacionadas con la anemia post destete.

Materiales y métodos: Se seleccionó un lechón pequeño, mediano y grande (N = 1095) por camada 1 a 2 días antes del destete de 20 granjas porcinas de Ontario (Canadá). Se recolectaron suero, muestras de sangre completa y pesos corporales. Tres semanas después, se tomó un segundo peso corporal y muestra de sangre de los mismos cerdos. Se analizaron la hemoglobina (Hb) y otros parámetros de sangre para evaluar el nivel de hierro y las asociaciones con el desempeño post destete. Se recolectaron los protocolos de suplementación de hierro y las concentraciones de ZnO en el alimento de área de destete.

Resultados: Se encontraron cerdos deficientes en hierro y anémicos al destete en la mayoría de las granjas participantes. Los cerdos que habían estado anémicos al destete fueron 0.82 kg más ligeros 3 semanas post destete que los lechones que tuvieron valores de Hb normales al destete ($P < .05$). Los lechones más grandes al destete presentaron hierro de suero y parámetros de células rojas, más bajos, y una capacidad de unión de hierro total más alta que lechones más pequeños (todos $P < .05$). Hubo más cerdos anémicos 3 semanas post destete que al destete ($P < .05$), y la prevalencia de anemia se asoció con altas concentraciones de ZnO ($P < .05$).

Implicaciones: Los protocolos de suplementación de hierro utilizados en los hatos de estudio fueron inadecuados para prevenir la deficiencia de hierro, particularmente en los cerdos más grandes. Los cerdos anémicos al destete tuvieron índices de crecimiento más lentos en el área de destete. El consumo de alimentos iniciadores en el área de destete que contenían altas concentraciones de ZnO se asocia con la anemia post destete.

Résumé - Étude sur la déficience en fer et l'anémie chez les porcelets et les effets du statut en fer au sevrage sur les performances post-sevrage

Objectifs: Déterminer le statut en fer de porcs au sevrage et les effets sur les

AMP, RMF, TLOS: Department of Population Medicine, University of Guelph, Ontario, Canada.

JCSH: Department of Large Animal Clinical Sciences, University of Saskatchewan, Saskatoon, Saskatchewan, Canada.

Corresponding author: Amanda Perri, Department of Population Medicine, University of Guelph, Guelph, ON N1G 2W1, Canada; Tel: 519-824-4120, ext 54854; E-mail: amperri@uoguelph.ca.

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

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performances post-sevrage, et déterminer si des concentrations élevées en oxyde de zinc (ZnO) dans la ration sont associées à de l'anémie en post-sevrage.

Matériels et méthodes: Un porcelet de petite, moyenne, et grande taille (N = 1095) ont été sélectionnés par portée 1 à 2 jours avant le sevrage de 20 fermes porcines en Ontario (Canada). Des échantillons de sérum et de sang entier ont été obtenus et le poids corporel noté. Trois semaines plus tard, à partir des mêmes animaux, on procéda à une deuxième série de prélèvements sanguins et de prise de poids. Le taux d'hémoglobine (Hb) et d'autres paramètres sanguins ont été analysés pour évaluer le statut en fer et les associations avec les performances post-sevrage. Les protocoles de supplémentation en fer et les concentrations de ZnO dans les rations en pouponnière ont été obtenus.

Résultats: Des porcs anémiques et avec des déficiences en fer au moment du sevrage étaient présents dans la plupart des fermes participantes. Les porcs qui avaient été anémiques au sevrage étaient plus léger de 0,82 kg 3 semaines post-sevrage que des porcelets qui avaient des valeurs normales de Hb au sevrage ($P < 0,05$). Les porcelets plus gros au sevrage avaient des paramètres érythrocytaires et une quantité de fer sérique inférieurs, et une capacité de liaison du fer total plus élevée que les plus petits porcelets (tous les $P < 0,05$). Plus de porcs étaient anémiques 3 semaines post-sevrage qu'au moment du sevrage ($P < 0,05$), et la prévalence d'anémie était associée avec des concentrations élevées de ZnO ($P < 0,05$).

Implications: Les protocoles de supplémentation en fer utilisés dans les troupeaux étudiés étaient inadéquats pour prévenir une déficience en fer, particulièrement chez les porcs plus gros. Les porcs anémiques au sevrage avaient des taux de croissance plus lents dans la pouponnière. La consommation de rations en pouponnière contenant des concentrations élevées en ZnO est associée à de l'anémie en post-sevrage.

It is well established that insufficient intake of iron in suckling pigs results in iron deficiency or anemia, where the concentration of hemoglobin (Hb) and the number and size of red blood cells (RBCs) decline below the normal range.[1] The suckling pig, regardless of breed, is susceptible to iron deficiency, anemia, or both.[2] The pig is born with limited iron, having a total body store of approximately 50 mg of iron, mostly incorporated in hemoglobin.[3] Sow milk is a poor source of iron, providing piglets with only 1 mg of iron a day.[3] Pigs lack access to soil, a rich source of iron,[3] due to confinement rearing indoors, and the modern pig has been selected for rapid growth. In the first week of life, piglets double their weight and increase their plasma volume by 30%, thereby diluting the concentration of Hb.[4] The daily iron requirement for piglets is approximately 7 mg, and, therefore, the limited iron that piglets are born with is inadequate in preventing iron deficiency and anemia, since these body stores dilute very rapidly.[3,5-8]

Piglets require exogenous iron supplementation within the first week of life to compensate for their limited iron and to prevent iron deficiency and anemia. It is commonly recommended to administer a 200-mg intramuscular (IM) injection of iron dextran within the first 3 days of life. Although oral iron is sometimes used, parenteral administration of iron is the most common method of iron supplementation for pigs on commercial swine farms.[9,10] A Hb concentration below 110 g per L is indicative of iron deficiency, and a Hb concentration below 90 g per L is indicative of anemia.[11,12] Iron deficiency occurs when there is a reduction (or usage without replacement) in the total content of iron in the body.[13] When there is a lack of iron in the body, nutrient requirements are not met. During the early stages, clinical signs such as anemia may not be apparent, whereas anemia occurs when iron deficiency is severe and causes a reduction in erythropoiesis.[13]

Iron supplementation is performed on a routine basis on commercial farms; however, the iron status of piglets is seldom evaluated. With updated management practices and modern genetic lines, sows farrow larger litters and piglets grow at an even greater rate than in previous decades.[14,15] Therefore, it is imperative to reassess whether the routine iron supplementation protocols used today on commercial swine farms are still adequate to prevent iron deficiency and anemia in modern piglets. This could have animal-health and economic implications, as piglets that have inadequate iron stores may develop a suppressed immune system, resulting in an impaired ability to resist infectious and parasitic diseases, and have a slower growth rate and increased morbidity and mortality.[9]

Iron deficiency, anemia, or both may occur on commercial swine farms because of husbandry errors, ie, inadequate dosing or timing of administration, or it may be that modern piglets require a higher dosage of iron during routine iron supplementation procedures. Also, common management practices, such as use of high concentrations of zinc oxide in the feed (> 2000 mg per kg) to control *Escherichia coli* diarrhea in newly weaned pigs, may decrease iron absorption from the feed. Copper, iron, and zinc are trace minerals that have similar physical and chemical properties. When there is an imbalance in one of these minerals, there may be an antagonistic effect on the nutritional availability of another mineral.[16] Thus, the use of high concentrations of zinc oxide in feed (> 500 mg per kg) may alter absorption of iron.

The objectives of this epidemiological study were to determine if iron deficiencies or anemia are present in pigs at weaning and if they affect post-weaning performance, and to determine if iron deficiency or anemia persists in the nursery stage and, if so, whether high concentrations of zinc oxide added to starter feeds is associated with the occurrence of post-weaning anemia.

Materials and methods

The Animal Care Committee at the University of Guelph, which follows the guidelines of the Canadian Council for Animal Care, reviewed and approved this study.

Study design and sampling

Twenty swine farms from 10 counties across southern Ontario were enrolled. The farms were sampled to represent a wide variety of production types, management practices, and sow-herd sizes. A questionnaire was administered at each farm to collect information regarding iron supplementation practices, including the age of piglet at the time of administration, dose, and type of iron supplementation product(s) used. The questionnaire also captured farm-management information such as the size of the sow herd, weaning age, and pig flow.

Each farm was visited twice. At the first visit, 1 to 2 days prior to weaning, litters were systematically selected starting at the first crate in the farrowing room until a maximum of 20 litters per farm was reached. Selection was based on visual assessment of three piglets per litter, including one large,

one medium, and one small piglet. Pigs were excluded if they exhibited physical abnormalities such as an abscess or hernia, or if they were lame or unthrifty, ie, thin body condition. Piglets were selected in this manner to obtain a balanced sample of different-sized piglets to enable assessment of iron status by body size. Each selected piglet was individually ear tagged and weighed. Blood samples were collected from each piglet via the orbital sinus technique using a Monoject standard hypodermic needle, 16-gauge × 1" (Covidien, Mansfield, Massachusetts). Blood was collected in 8.5-mL tubes (BD Vacutainer; BD, Franklin Lakes, New Jersey), and whole blood samples were collected in 6-mL tubes containing ethylenediamine tetraacetic acid (EDTA) (BD Vacutainer; BD). At the second visit, 3 weeks after the first visit, the same pigs were weighed and whole blood samples were collected using the techniques described. In order to evaluate the prevalence of iron deficiency and anemia in piglets at weaning, a classification for Hb status was determined a priori, based on current classifications found in the literature. Normal iron status was defined as a Hb value of > 110 g per L, iron deficiency was defined as a Hb value of > 90 g per L but ≤ 110 g per L, and anemia was defined as a Hb value of ≤ 90 g per L.[11,12] The prevalence of iron deficiency and anemia in piglets was determined for each farm.

Hemoglobin measurement

Hemoglobin values were analyzed using two methods. On the initial sampling, 1 to 2 days prior to weaning, the whole blood samples collected from each of the piglets were analysed at the Animal Health Laboratory (AHL) at the University of Guelph using the ADVIA 2120 per 2120i Hematology system (Siemens Healthcare Diagnostics, Deerfield, Illinois) as per standard protocols. Briefly, the blood sample and the ADVIA 2120 HGB reagent are mixed together in the Hb chamber of a colorimeter. The Hb reaction involves two steps: RBCs are lysed to release Hb, and heme iron found in Hb is oxidized from the ferrous to the ferric state, and then combined with cyanide in the ADVIA 2120 HGB reagent to form the product.[17] Optical readings are obtained colorimetrically at 546 nm.[17]

The second Hb measurement occurred 3 weeks after the initial visit, when the pigs were in the nursery. At this time, Hb in whole blood samples was measured using an automated hematology handheld instrument (STAT-Site M Hgb meter, Boerne, Texas). The STAT-Site M Hgb meter contains a plastic card with reagent pads for determining the concentration of Hb. This device provides measurements of blood Hb content within 30 seconds after 15 μL of blood has been applied to the test strip. The amount of color produced from azide-methemoglobin is proportional to the concentration of Hb in the sample.[18]

Hematology measurements

The red blood cell (RBC) count, hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) in whole blood samples collected at the first visit were analyzed by the AHL using the ADVIA 2120 per 2120i Hematology System (Siemens Healthcare Diagnostics). All of these indicators of iron status were analyzed from a single optical cytometer after dilution of the samples with ADIVA 2120 RBC reagent. As the reaction mixture moves towards the flowcell in the optical cytometer, a laser strikes the cells and generates electronic scatter signals to measure the size, volume, and internal characteristics of the cells.

To determine serum iron and total iron binding capacity (TIBC), the same whole blood samples were also analyzed by the AHL using a Roche per Hitachi cobas 6000 c501 analyzer (Roche Diagnostics International Ltd, Rotkreuz, Switzerland). Briefly, iron is released from transferrin under acidic conditions. Ascorbate decreases the number of released Fe^{+++} ions to Fe^{++} ions, which then react with the iron reagent ferrozine.[17] This reaction forms a colored complex in which TIBC is measured photometrically and is proportional to the color intensity.[13]

Zinc oxide levels in the feed

Feed tags were collected at each farm to evaluate the concentration of zinc oxide in the first-phase nursery diets. The Canadian Food Inspection Agency (CFIA) and Feeds Regulations (1983) state that the maximum zinc oxide concentration in feed is 500 mg per kg for nursery pigs, without a request from the producer to increase the concentration.[19] The CFIA also states that the actual amount of zinc oxide in mg per kg of the feed must be present on the label. All participating farms fed diets meeting daily zinc requirements and higher. Zinc oxide concentration in the feed was categorized: nutritional dose (≤ 500 mg per kg), high dose (2000 to 3000 mg per kg), and very high dose (> 3000 mg per kg).

Statistical analysis

All data were entered into an Excel spreadsheet (Microsoft Corporation, Redmond, Washington) and then imported into Stata 12 Intercooled for Windows XP (Statacorp LP, College Station, Texas) for analyses. The association between Hb status (g per L) at weaning and subsequent nursery growth performance (measured as the 3-weeks-post-weaning weight) was analyzed using mixed linear regression. The dependent variable in the model was the 3-weeks-post-weaning weight. Extraneous variables that were used in the final model include parity of the sow, age at weaning, weaning weight, and Hb status at weaning. The model was built using forward stepwise criteria and designed parsimoniously. Hemoglobin status was categorized as follows: normal Hb (> 110 g per L), iron deficient (> 90 g per L but ≤ 110 g per L) or anemic (≤ 90 g per L). Dam (sow) parity was categorized as parity 1 (gilt), parity 2 to 5, and parity > 5. The age of the piglet when the iron supplementation was administered was categorized as ≤ 1 day of age, 2 to 4 days of age, and 5 to 7 days of age.

All extraneous variables were initially screened for univariable associations using linear regression and considering farm as a random effect. Univariable associations with a liberal P value of < .2 were considered for the final model. Linearity of continuous predictor variables with 3-weeks-post-weaning weight as the outcome variable was assessed using two methods: visually using a lowess smoother (smoothed locally weighted scatter plot), and using a quadratic term. Confounding was assessed throughout model building. A confounding variable was defined as a variable whose removal from the model changed the coefficient of any predictor variable by greater than 20%. Two-way interactions were generated between all extraneous variables in the initial model and were included in the final model only if they were statistically significant ($P < .05$). Extraneous variables were included in the final model if variables had a $P < .2$ in univariable analysis and then a $P < .05$ in the final model, if the variable was a confounder, or if the variable was part of a statistically significant interaction ($P < .05$). These variables were then assessed for collinearity

using Pearson correlation analysis. Potential outliers, influential points, and the model assumptions were assessed graphically.

To assess the association between various iron indicators and weaning weight (category), eight separate linear mixed models were created. Each iron indicator (RBC, Hb, HCT, MCV, MCH, MCHC, serum iron, and TIBC) was modeled separately as the dependent variable and in each of the models, the association between the size of the piglet at weaning (small, medium, or large) and each iron indicator was evaluated. In these models, sex of the piglet, parity of the dam, age at weaning, weaning weight, and the type of iron administered were modeled as fixed effects, and farm was modeled as a random effect using the methods described for analysis involving Hb.

To explore the association between the zinc oxide content in feed and anemia at 3 weeks post weaning, a mixed logistic regression model was built. For this model, iron status was dichotomized as anemic if the piglet had a Hb concentration ≤ 90 g per L or normal if the piglet had a Hb concentration > 90 g per L. Iron status at weaning, the type of iron administered, and age at weaning were modeled as fixed effects. Zinc oxide concentration in the feed was categorized as nutritional dose, high dose, or very high dose, as described. Confounding was assessed throughout model building as described. Potential outliers and influential points were evaluated graphically, and model diagnostics were performed.

Results

The farms enrolled varied in size from 112 to 1500 sows, with the majority of farms being farrow-to-finish and four being farrow-to-wean. For the farrow-to-wean farms, the initial sampling visit occurred at each of the four sites, and then the second sampling visit occurred at each of the farms' off-site nurseries. A total of 1095 pigs were sampled, with a range of 13 to 20 litters per farm. All male piglets enrolled in this study had been castrated. Farm-specific demographics, iron supplementation protocols, and mean growth performance values, including piglet weaning weight, 3-weeks-post-weaning weight, and average daily gain (ADG) for each farm, are presented in Table 1. Of the 20 farms sampled, 60% (12 of 20) mixed their iron product with other pharmaceutical products such as meloxicam or penicillin.

The mean age (± standard deviation) at which piglets were initially sampled (1 to 2 days prior to weaning) was 21.8 ± 4.2 days. The mean weight of pigs at initial sampling (1 to 2 days prior to weaning) was 6.4 ± 1.8 kg. The mean weight of pigs in the small (n = 365), medium (n = 365), and large (n = 365) weight categories were 5.2 ± 1.5 kg, 6.5 ± 1.4 kg, and 7.5 ± 1.6 kg, respectively. The prevalence of iron deficiency and anemia at 1 to 2 days prior to weaning and at 3 weeks post weaning for individual farms are presented in Table 1. The within-herd prevalence of iron deficiency and anemia at weaning ranged between 0% and 61% and 0% and 46%, respectively. The between-herd prevalences of iron deficiency and anemia at weaning were 28% and 6%, respectively. Nineteen of the 20 farms (95%) had piglets with low Hb values (iron deficient or anemic or both) at weaning. Upon sampling at 3 weeks post weaning, the within-herd prevalence for iron deficiency and anemia ranged between 29% and 74% and 6% and 32%, respectively. The between-herd prevalences of iron deficiency and anemia at this time were 43% and 18%, respectively. From the initial sampling day to 3 weeks post weaning, 72 pigs (6.6%) were lost from the study. The reasons for piglet loss were not recorded, but pig loss was evenly distributed among the farms. Of the 72 pigs that were missing, 58% had normal Hb values, 35% were iron deficient, and 7% were anemic at weaning.

The associations between piglet weight categories at weaning and iron indicators are presented in Table 2. Medium-sized piglets at weaning had a Hb concentration 2.7 g per L higher than larger pigs at weaning ($P < .01$). Smaller-sized pigs at weaning had a Hb concentration 3.4 g per L greater than large pigs at weaning ($P < .001$). Hemoglobin status at weaning did not differ between small and medium weight categories.

The eight models illustrating the associations between various iron indicators with bodyweight category at weaning are presented in Table 3. The mean values for various iron indicators, by piglet weight category, can be found in Table 4. Piglets from the large weight category had lower Hb, serum iron, HCT, MCV, MCH, and MCHC values than did piglets in the small and medium weight categories ($P < .05$). Total iron binding capacity values were higher in the large-sized piglets than in the small and medium-sized piglets ($P < .01$). There was no statistical difference found between each of the weight categories and RBC counts.

The final model illustrating the association between Hb status at weaning and 3-weeks-post-weaning weight is presented in Table 4. No significant interactions or confounders were identified. The final model revealed that anemic piglets at weaning had a 0.82 kg lower 3-weeks-post-weaning weight than did piglets with normal Hb values at weaning ($P < .01$). Also, anemic pigs at weaning were on average 0.69 kg lighter in weight at 3 weeks post weaning than pigs that were classified as iron deficient at weaning ($P < .05$). There was no statistical difference in 3-weeks-post-weaning weight when comparing iron-deficient pigs at weaning with pigs with a normal Hb status. Piglets from sows whose parities ranged from 2 to 5 and from sows of parities > 5 had higher 3-weeks-post-weaning weights than did piglets from gilts ($P < .05$). There was no difference in the 3-weeks-post-weaning weights between piglets administered an iron dextran injection or a gleptoferron injection. Piglets weaned at an older age had a 0.12-kg higher weight at 3 weeks post weaning ($P < .001$).

The zinc oxide content in feed, collected from feed tags, ranged between 250 and 7000 mg per kg on the farms. The logistic regression model created to explore the association between the zinc oxide content in feed and anemia at 3 weeks post weaning is presented in Table 5. The odds of nursery pigs being anemic was 3.4 times greater for pigs consuming high doses of zinc oxide in feed than in those consuming a nutritional dose of zinc oxide in feed ($P < .05$). The odds of nursery pigs being anemic was 4.1 times greater for those consuming starter feeds containing very high concentrations of zinc oxide than for those consuming a nutritional dose of zinc oxide ($P < .05$). There was no difference in the odds of anemia in pigs fed very high doses of zinc oxide, compared to pigs fed a high dose of zinc oxide ($P > .05$). The type of iron administered was a confounder in this model. This is likely because anemia was classified differently (dichotomized) in this model, but was classified categorically in the main model, and because the majority of farms used iron dextran, hence this variable was included in the model.

Discussion

Despite routine iron supplementation during the first week of life, pigs with low Hb values were identified at weaning on almost all farms, and, surprisingly, the prevalence of anemic pigs was greater 3 weeks after weaning.

Table 1: Summary of farm production parameters, iron supplementation protocols, and iron status of piglets from 20 commercial swine farms in Ontario (Canada) at weaning and 3 weeks post weaning*

Farm (no. sows)	No. pigs at weaning (no. litters)	Age (days)†		Mean weight (kg)‡		Iron status§						ADG (kg)
						% at weaning (n)			% 3 weeks post weaning (n)			
		Iron	Weaning	Weaning	Post-weaning	Normal	Deficient	Anemic	Normal	Deficient	Anemic	
1 (600)	57 (19)	< 1	21.4 (± 2.2)	6.7 (± 1.7)	12.0 (± 2.9)	35 (20)	61 (35)	4 (2)	63 (35)	30 (17)	7 (4)	0.25 (± 0.06)
2 (1400)	60 (20)	2-4	19.2 (± 0.9)	6.3 (± 1.4)	13.4 (± 2.4)	63 (38)	37 (22)	0 (0)	ND	ND	ND	0.34 (± 0.08)
3 (500)	60 (20)	5-7	27.9 (± 2.8)	8.9 (± 1.9)	17.4 (± 2.9)	72 (43)	27 (16)	1 (1)	62 (37)	28 (17)	10 (6)	0.40 (± 0.09)
4 (300)	60 (20)	5-7	20.9 (± 1.7)	5.7 (± 1.3)	11.5 (± 2.3)	80 (48)	12 (7)	8 (5)	20 (12)	57 (34)	23 (14)	0.27 (± 0.06)
5 (250)	51 (17)	5-7	24.5 (± 1.3)	6.6 (± 1.2)	12.7 (± 2.7)	37 (19)	39 (20)	24 (12)	19 (9)	75 (35)	6 (3)	0.29 (± 0.09)
6 (112)	48 (16)	5-7	26.2 (± 2.7)	6.4 (± 1.4)	11.5 (± 3.1)	6 (3)	48 (23)	46 (22)	28 (13)	64 (30)	8 (4)	0.23 (± 0.09)
7 (1000)	60 (20)	< 1	21.0 (± 1.7)	6.5 (± 1.4)	12.6 (± 1.9)	30 (18)	58 (35)	12 (7)	59 (35)	29 (17)	12 (7)	0.29 (± 0.06)
8 (850)	60 (20)	2-4	18.7 (± 1.3)	5.9 (± 1.4)	9.3 (± 2.6)	70 (42)	25 (15)	5 (3)	35 (21)	33 (20)	32 (19)	0.16 (± 0.08)
9 (140)	39 (13)	< 1	29.5 (± 3.2)	5.2 (± 1.3)	7.3 (± 1.7)	59 (23)	33 (13)	8 (3)	26 (10)	44 (17)	30 (12)	0.10 (± 0.06)
10 (1250)	60 (20)	2-4	25.3 (± 1.6)	7.2 (± 1.5)	14.4 (± 2.8)	83 (50)	17 (10)	0 (0)	27 (16)	42 (25)	31 (18)	0.36 (± 0.08)
11 (130)	39 (13)	2-4	25.5 (± 3.5)	7.7 (± 1.8)	14.5 (± 3.2)	100 (39)	0 (0)	0 (0)	28 (11)	46 (18)	26 (10)	0.31 (± 0.16)
12 (640)	60 (20)	2-4	17.5 (± 1.0)	6.0 (± 1.2)	11.0 (± 2.2)	80 (48)	20 (12)	0 (0)	13 (8)	51 (30)	36 (21)	0.24 (± 0.08)

Table 1 continued on page 15

Different reference limits are reported in the literature regarding how low Hb concentration can be before anemia is diagnosed.[10-12] Wei et al[10] suggest that a Hb concentration above 100 g per L is considered normal and that a Hb concentration below 60 g per L indicates severe anemia. Anemia has also been defined as a Hb concentration below 80 g per L.[9] A recent paper by Bhattarai and Nielsen[12] used a Hb concentration below 110 g per L as indicative of iron deficiency and a Hb concentration below 90 g per L as indicative of anemia. Being able to distinguish low iron prior to evidence of clinical anemia is a useful concept for practitioners who are monitoring the effectiveness of an iron supplementation program, and for this reason we chose to use the Bhattarai and Nielsen[12] categories to assess Hb concentrations. Since a slightly higher threshold was chosen for defining iron deficiency and anemia, compared to that in other published material, this may partly be the reason why the prevalences were high.

Hemoglobin is commonly used as a measurement of iron status, because 80% to 90% of the iron present in the suckling piglet is used in forming Hb.[20] Hemoglobin is an important protein involved in cellular metabolism, as it transports oxygen from the lungs to other body tissues, and transports carbon dioxide back to the lungs for expulsion via the respiratory tract.[4,14] The greater the concentration of Hb per given unit of blood, the greater the amount of oxygen that can be carried in blood.[21] Iron deficient and anemic pigs have fewer RBCs containing less Hb, compared to piglets with normal Hb levels.[4] Along with Hb, other iron indicators, such as serum iron levels and TIBC, are also important for assessing iron status in swine. Serum iron measures the amount of iron circulating in the blood bound to transferrin, an important protein that binds and transports iron in blood. Total iron binding capacity measures the blood's capacity to bind with transferrin.

Although Hb status is the most frequently used parameter for evaluating iron deficiency and anemia in swine, it is possible that other blood parameters may be more sensitive in detecting the early stages of iron deficiency.[22] For instance, Bhattarai and Nielsen[12] were not able to find a difference in hemoglobin concentration between various piglet sizes, but found that large pigs had lower serum iron and higher TIBC than other pigs, indicating that iron is utilized faster in bigger piglets, making them prone to iron deficiency. Bhattarai and Nielsen[12]

Table 1 continued

Farm (no. sows)	No. pigs at weaning (no. litters)	Age (days)†		Mean weight (kg)‡		Iron status§						ADG (kg)
						% at weaning (n)			% 3 weeks post weaning (n)			
		Iron	Weaning	Weaning	Post-weaning	Normal	Deficient	Anemic	Normal	Deficient	Anemic	
13 (1500)	60 (20)	2-4	19.9 (± 0.7)	5.6 (± 1.4)	11.1 (± 2.1)	97 (58)	3 (2)	0 (0)	28 (17)	45 (27)	27 (16)	0.26 (± 0.05)
14 (600)	57 (19)	2-4	17.1 (± 1.3)	5.2 (± 1.2)	8.6 (± 1.6)	77 (44)	23 (13)	0 (0)	51 (29)	37 (21)	12 (7)	0.17 (± 0.04)
15 (535)	60 (20)	< 1	21.1 (± 2.7)	6.8 (± 1.7)	13.3 (± 3.0)	70 (42)	30 (18)	0 (0)	48 (29)	32 (19)	20 (12)	0.22 (± 0.06)
16 (250)	60 (20)	< 1	18.0 (± 1.7)	5.1 (± 1.2)	8.7 (± 2.1)	92 (55)	8 (5)	0 (0)	34 (20)	53 (32)	13 (8)	0.16 (± 0.06)
17 (85)	42 (14)	2-4	18.3 (± 4.3)	5.3 (± 1.5)	12.8 (± 3.4)	48 (20)	50 (21)	2 (1)	37 (15)	49 (20)	14 (6)	0.36 (± 0.10)
18 (420)	60 (20)	2-4	20.8 (± 1.4)	6.4 (± 1.4)	11.2 (± 1.7)	98 (59)	2 (1)	0 (0)	40 (24)	35 (21)	25 (15)	0.23 (± 0.06)
19 (262)	42 (14)	2-4	21.7 (± 4.0)	6.0 (± 1.2)	11.6 (± 2.1)	31 (13)	52 (22)	17 (7)	37 (15)	56 (23)	7 (3)	0.26 (± 0.07)
20 (650)	60 (20)	5-7	26.4 (± 2.8)	8.4 (± 1.5)	14.8 (± 2.4)	77 (46)	23 (14)	0 (0)	75 (44)	24 (14)	1 (1)	0.30 (± 0.07)
Total (573)	**54.8 (18.2)**	**NA**	**21.8 (± 2.1)**	**6.4 (± 1.4)**	**12.0 (± 2.4)**	**65.2 (728)**	**28.4 (304)**	**6.4 (63)**	**38.3 (400)**	**43.7 (437)**	**18.0 (186)**	**0.26 (± 0.07)**

* Farms 2, 7, 12, and 13 were farrow-to-wean farms: all other were farrow-to-finish farms. Farms 1, 6, 8, 14, and 20 used a gleptoferron product as an iron supplement; all other farms used an iron dextran product.
† Age at iron administration and mean age at weaning (± SD).
‡ Body weight (mean ± SD) at weaning and 3 weeks post weaning; ADG calculated by (3-weeks-post-weaning weight – weaning weight) ÷ no. of days between visits.
§ Normal, hemoglobin (Hb) > 110 g/L; iron deficient, Hb ≤ 110 g/L but > 90 g/L; anemic, Hb ≤ 90 g/L. At weaning, Hb was measured at the Animal Health Laboratory; at 3 weeks post weaning, Hb was measured using a hand-held meter (STAT-Site M Hgb meter; Boerne, Texas).
SD = standard deviation; ADG = average daily gain; NA = not applicable; ND = not done (lost samples).

also concluded that using Hb as a diagnostic tool may underestimate the iron requirements for young growing piglets. With this in mind, additional iron indicators were analyzed in this study. The results for serum iron, HCT, MCV, MCH, and MCHC in each pig weight category at weaning agreed with the results of the Hb measurements for assessing iron status in this study. Because Hb can be easily and inexpensively measured using hand-held instruments that can be used on farm, these results support the continued use of Hb to monitor iron status on pig farms.

All of the measurements for iron status used in the present study indicate that the larger piglets at weaning were more likely to be iron deficient than were the small and medium-sized piglets. The data from the current study also indicate that, on most farms, the traditional supplementation of 200 mg of parenteral iron is insufficient to meet the needs of the large and fast-growing piglets, and a higher dosage of iron or a second injection of iron at a later date during the suckling period may be required.

The prevalence of anemia and iron deficiency in pigs at weaning found in the current study is similar to the results from recent studies in various countries.[11] Walsh et al[23] found 30% of Ontario pigs to be anemic at weaning, when assessing Hb status on a single commercial swine farm. The current study confirms that identifying iron-deficient or anemic piglets at weaning is not uncommon on Ontario commercial pig farms. There are other possible reasons, in addition to greater nutritional requirements for fast growing pigs, which might explain why anemic and iron-deficient piglets are present at weaning. One possible reason is human error during administration of the iron supplementation, eg, some piglets are missed during the process or iron is given late. It is also possible that there could be injection-site leakage resulting in dose variation when iron is administered during processing, with some pigs thereby more at risk for anemia because they did not receive a full dose of iron product. In this study, on 60% of the participating farms, an iron product was used that had been mixed with penicillin or meloxicam. This is surprising because "mixing two or more medications in a syringe for delivery to animals is a form of compounding and is not permitted" according to the Canadian Quality Assurance program (CQA Producer Manual, Version 2.1, D4-6; 2007)[24] and Health Canada's policy on drug compounding in human and veterinary medicine (Policy on Manufacturing and Compounding Drug Products in Canada POL-0051; 2009).[25] To the authors' knowledge, it is unknown what effect, if any, compounding pharmaceuticals with iron products may have on the uptake

Table 2: Eight individual models illustrating the associations between various iron-status indicators and the body-weight category 1 to 2 days prior to weaning among 1095 piglets from 20 Ontario (Canada) commercial swine farms

Model*	Weight category†	Coefficient‡	SE	95% CI	*P*	Contrast medium versus small pigs coefficient	SE	95% CI	*P*
Hb (g/L)	Small	3.43	0.923	1.625, 5.243	< .001	-.734	0.923	-2.543, 1.076	.43
	Medium	2.70	0.923	0.891, 4.510	< .001				
Serum iron (umol/L)	Small	6.95	0.820	5.345, 8.560	< .001	-3.540	0.820	-5.149, 1.933	< .001
	Medium	3.41	0.820	1.803, 5.019	< .001				
Hematocrit (L/L)	Small	0.01	0.003	0.001, 0.013	.02	0.000	0.003	-0.006, 0.006	.98
	Medium	0.01	0.003	0.001, 0.013	.02				
Mean corpuscular volume (fL)	Small	2.25	0.371	1.521, 2.974	< .001	-0.969	0.371	-1.695, -0.242	.01
	Medium	1.28	0.371	0.552, 2.005	< .001				
Mean corpuscular Hb (pg)	Small	0.91	0.153	0.679, 1.148	< .001	-0.408	0.119	-0.643, -0.174	< .001
	Medium	0.51	0.182	0.271, 0.740	< .001				
Mean corpuscular Hb concentration (g/L)	Small	3.48	0.690	2.130, 4.835	< .001	-1.930	0.690	-3.283, -0.578	< .001
	Medium	1.55	0.690	0.200, 2.905	.02				
Total iron binding capacity (umol/L)	Small	-14.07	1.375	-16.763, -11.374	< .001	8.764	1.375	6.068, 11.459	< .001
	Medium	-5.31	1.375	-8.000, -2.609	<.001				
Red blood cells (10^{12}/L)	Small	-0.09	0.050	-0.183, 0.012	.09	0.082	0.050	-0.016, 0.179	.10
	Medium	-0.00	0.50	-0.101,0.094	.94				

* Mixed linear regression models using Stata 12 Intercooled XP for Windows (College Station, Texas). For all eight models, parity category of the dam, age at weaning, and weaning-weight category were modelled as fixed effects and farm was modelled as a random effect (coefficient not shown), with iron status indicator as the dependent variable.

† Weight categories include small (5.2 ± 1.5 kg), medium (6.5 ± 1.4 kg), and large (7.5 ± 1.6 kg) (referent) piglets selected.

‡ Coefficient represents the change in each iron analyte 1 to 2 days prior to weaning, comparing small and medium-sized piglets to large piglets, eg, smaller piglets 1 to 2 days prior to weaning have hemoglobin values 3.4 g/L higher than larger piglets 1 to 2 days prior to weaning.

SE = standard error; CI = confidence interval; Hb = hemoglobin.

of iron by the piglet. Nevertheless, if iron is mixed with other products there is a risk that one of the products settles and when the compounded product is drawn into a syringe the proportion of iron may not be the expected concentration, so that some piglets are underdosed while others receive a high dose.

The rapid growth rate of modern piglets is a concern because iron requirements are likely increased. The large-sized piglets in this study had lower Hb concentrations at weaning than did the small and medium-sized piglets. Jolliff and Mahan[14] also found that heavier piglet weaning weight was associated with lower Hb and HCT values. The reason for this may be explained by the fact that each piglet receives a fixed amount of iron from maternal stores.[6] Smaller piglets will have less blood volume, thus having a higher concentration of Hb for optimum synthesis. Larger piglets have a larger blood volume, therefore diluting Hb and increasing their iron requirements, making them more susceptible to iron deficiency and anemia. In this study, the larger pigs selected at weaning had lower Hb values than did the small and medium-sized pigs, indicating that a single 200-mg IM injection of either iron dextran or gleptoferron is not sufficient to prevent iron deficiency and anemia in some rapidly growing pigs.

The timing of the iron injection, specifically the age of the pig when iron is administered, is also important to consider when assessing Hb status at weaning. The producer from each participating farm completed a questionnaire and indicated the age at which iron was administered. However, in reality, there was likely minor variation. It is unknown how stringently the producers followed their own iron-supplementation protocol, since it may not always be possible to administer iron on the same day of age after every litter of piglets is born. This limitation of the study may have introduced some misclassification bias to the variable "day of iron administration." The literature indicates that parenteral iron supplementation within the first 3 to 4 days of age generally prevents anemia in suckling pigs.[6] In this study, the range of ages at which suckling pigs were administered iron was within the first 24 hours up until 7 days of age, and the majority of producers reported that they administered iron within 3 to 4 days of age.

Table 3: The mean (± SD) of various iron analytes in 1095 piglets sampled prior to weaning from 20 Ontario (Canada) commercial swine farms*

Analyte	Small pigs (n = 365)	Medium pigs (n = 365)	Large pigs (n = 365)	All pigs
Hb (g/L)	114.1 (± 15.34)	113.3 (± 15.75)	110.6 (± 15.38)	112.7 (± 15.55)
Serum iron (umol/L)	22.9 (± 11.73)	19.2 (± 12.94)	15.8 (± 11.80)	19.3 (± 12.50)
Hematocrit (L/L)	0.39 (± 0.05)	0.39 (± 0.05)	0.39 (± 0.05)	0.38 (± 0.50)
Mean corpuscular volume (fL)	66.7 (± 6.89)	65.6 (± 7.08)	64.4 (± 7.03)	65.6 (± 7.06)
Mean corpuscular Hb (pg)	19.8 (± 2.15)	19.3 (± 2.25)	18.8 (± 2.26)	19.3 (± 2.25)
Mean corpuscular Hb conc (g/L)	295.8 (± 12.46)	293.9 (± 11.35)	292.3 (± 12.44)	294.0 (± 12.17)
Total iron binding capacity (μmol/L)	74.4 (± 24.89)	83.3 (± 22.53)	88.5 (± 22.08)	82.1 (± 23.91)
Red blood cells (10^{12}/L)	5.8 (± 0.78)	5.9 (± 0.80)	5.9 (± 0.74)	5.9 (± 0.77)

* Mean age at weaning, 21.8 ± 4.2 days. Mean weights (± SD) at weaning: small pigs, 5.2 ± 1.5 kg; medium pigs, 6.5 ± 1.4 kg; large pigs, 7.5 ± 1.6 kg.

Hb = hemoglobin; SD = standard deviation; conc = concentration.

Table 4: The final model* illustrating the effect of iron deficiency, anemia, weight and age at weaning, and parity at weaning on weight 3 weeks post weaning in pigs from 20 Ontario (Canada) commercial swine farms.

Variable	Coefficient	SE	95% CI	*P*
Hemoglobin status				
Iron deficient	-0.13	0.129	-0.385, 0.120	.30
Anemic	-0.82	0.259	-1.327, -0.313	< .01
Weight at weaning	1.25	0.037	1.177, 1.323	< .001
Parity				
2-5	0.34	0.157	0.035, 0.651	.03
> 5	0.50	0.187	0.137, 0.869	< .01
Age at weaning	0.12	0.023	0.072, 0.163	< .001

* Mixed linear regression with farm modeled as random effect. Coefficients represent the change in weight (kg) 3 weeks post weaning if the variable is increased by one unit or compared to its referent category, eg, in piglets that were anemic (defined in Table 1) 1-2 days prior to weaning, 3-weeks-post-weaning weight was 0.82 kg lower than that in piglets with normal Hb. When a statistical contrast was conducted, anemic pigs at weaning had a 0.69-kg lower 3-weeks-post-weaning weight on average than that of pigs classified at weaning as iron deficient (defined in Table 1) ($P < .05$). Referent parity was parity 1 (gilts). Mean age at weaning was 21.8 ± 4.2 days.

Hb = hemoglobin; SE = standard error; CI = confidence interval.

This may explain why this study did not find an association with timing of administration of iron supplementation, since the majority of farms administered iron products by the time pigs were 7 days of age.

The type of iron administered and the dose are important considerations when assessing Hb at weaning. Among the 20 participating farms, all farms supplemented their piglets with an IM injection of either a gleptoferron or iron dextran product. The majority of farms in this study used iron dextran. However, no difference was found in the Hb status of pigs when farms using iron dextran and gleptoferron were compared. This finding is consistent with other studies that have reported no difference between gleptoferron and iron dextran in preventing iron deficiency and anemia.[26,27] This suggests that iron from both gleptoferron and iron dextran is utilized with comparable efficacy for hemoglobin synthesis and iron storage in young growing pigs. On most farms, pigs have access to creep feed containing iron. It was not possible in the current study to determine whether the intake of creep feed contributed to the piglet's iron status. It was thought that because most piglets were weaned at approximately 3 weeks of age, only a small amount of feed would have been consumed and may not have been a factor in meeting the piglet's iron needs.

A different test method was used to assess Hb status at 3 weeks post weaning than was used to assess the suckling piglets. Testing method is confounded with differences between Hb at weaning and 3 weeks post weaning. Hemoglobin status has been evaluated using a handheld device on farm.[28-30] During the second visit to each participating farm, Hb measurements were evaluated using a STAT-Site M Hgb handheld meter.

Table 5: Model* assessing the association of zinc oxide concentration in feed and odds of anemia in piglets 3 weeks post weaning 20 Ontario (Canada) commercial swine farms.

Variable	OR	SE	95% CI	*P*
Zinc oxide concentration				
High dose	3.4	1.974	1.114-10.595	.03
Very high dose	4.1	2.551	1.206-13.889	.02
Hemoglobin at weaning	1.0	0.006	0.971-0.996	< .01
Weight at weaning	0.9	0.052	0.822-1.025	.13
Iron administered	2.9	1.454	1.086-7.749	.03

* Mixed logistic regression. OR represents the odds of anemia, eg, the odds of nursery pigs being anemic was 3.4 times greater for pigs consuming high doses of zinc oxide in feed than for those consuming a nutritional dose of zinc oxide in feed. The referent was the nutritional dose (≤ 500 mg/kg of feed), the high dose was 2000-3000 mg/kg of feed, and the very high dose was > 3000 mg/kg of feed). The referent for the type of iron administered was gleptoferron.

OR = odds ratio; SE = standard error; CI = confidence interval.

This convenient handheld meter can be utilized while on farm to assess analytical results more rapidly than by submitting samples to a laboratory service. A limitation to using this particular handheld device is that it has not been used to assess Hb concentration in swine. However, this device has been tested on humans and has a 0.93 correlation coefficient when compared to standard laboratory testing.[31] Another handheld meter, the HemoCue, is a similar device that has been used to assess Hb measurements in both humans and swine.[28,29] This device measures Hb content via the conjugation of free Hb to azidemethemoglobin, photometrically measured at 570 nm, whereas the STAT-Site M Hgb meter uses the same method but measures photometrically at 565 nm.[29,30] Kutter et al[28] reported good agreement after assessing the HemoCue meter with standardized Hb laboratory measurements. The sensitivity and specificity of the HemoCue meter were 97% and 100%, respectively.[31]

Similar to suckling piglets, nursery pigs also grow rapidly, resulting in a rapid increase in blood volume and a high nutritional iron requirement.[32] The decrease in Hb concentrations when the nursery pigs were tested using the STAT-Site M Hgb meter suggests that Hb synthesis may not increase proportionally as the rapidly growing nursery pigs increase in weight and blood volume.[14] There are several possible reasons why iron deficiency and anemia continued 3 weeks post weaning. Firstly, the types of iron used in the nursery diets may have varied among farms, some being more readily absorbed than others. Although the iron concentrations varied in the nursery diets, all were well over National Research Council requirements. The stress of weaning is also associated with a reduction in feed consumption, which may play a role in iron deficiency and anemia post weaning.[33] In a previous study, it was suggested that intestinal regulation of iron absorption might not be entirely functional within the first few weeks post weaning.[34] Divalent metal transporter 1 (DMT1) is an important membrane protein that plays a key role in intestinal iron absorption as well as iron transport.[35] Hansen et al[32] found that mRNA transcript levels of DMT1 are not up-regulated in pigs until they reach 26 to 27 days of age. Therefore, there is evidence that iron absorption and transport are not regulated in pigs until they are in the nursery. In addition, iron absorption is controlled by hepcidin antimicrobial peptide (HAMP), which is derived from the liver and is produced in response to high concentrations of iron in the feed.[34] HAMP binds to ferroportin (an iron exporter) on cell surfaces and degrades the cells.[36] Hansen et al[32] also concluded that HAMP-mediated iron homeostasis is likely not fully functional in newly weaned pigs and that these pigs are not able to properly react to changes in dietary iron. In the current study, piglets that were anemic grew more slowly. This might lead to economic issues in the future if mortality and morbidity rates are elevated due to underlying anemia.

Prevention of iron deficiency in piglets at the time of weaning needs to be investigated. There is a danger of iron toxicosis and a concern of increasing bacteremia if the dosage of iron administered within the first few days of life is higher than that recommended by the manufacturer. A second injection close to or at the time of weaning may be a good way to provide sufficient iron to the pig to meet the nutritional demands of the early weanling stage without increasing the risk of toxicity, but a second injection is associated with an increased labor cost. There may be an economic benefit to adding a second injection to the overall benefit of piglet health and performance that might outweigh the cost of labor, but this needs to be assessed on individual farms and warrants further study. However, Peters and Mahan[37] found that suckling pigs that were injected with 200 mg of iron at birth and then a second time at weaning did not respond to the additional injection, and thus this also needs to be further investigated.

Iron deficiency and anemia was associated with lower growth rates (poor nursery performance) in this study. Pigs that were iron deficient and anemic at weaning had mean 3-week nursery body weights lower than pigs with normal Hb levels at weaning, and this is consistent with other reports.[38,39] Schrama et al[38] found that piglets with low Hb values had lower ADG than piglets with higher Hb levels. Gentry et al[39] also found that pigs with higher Hb levels at weaning had greater ADG and higher feed intake post weaning. These results are comparable to the current study, since 3-weeks-post-weaning weight was positively associated with Hb status at weaning.

In order to investigate the effects of Hb status on post-weaning performance, various farm management protocols were accounted for in the analyses. Piglet weaning weight and age were controlled for because they are both

significant contributors to 3-week growth performance in nursery pigs. Piglet weight at weaning was positively associated with 3-weeks-post-weaning weight in the nursery barn, which is consistent with previous studies.[40,41] Piglets weaned at an older age reached a greater 3-weeks-post-weaning weight than did piglets weaned at an earlier age.

Both litter size and sex were not included in the final model because these variables had no significant association with 3-weeks-post-weaning weight in univariable analysis. Sow parity was included in the final model, as there was a statistically significant association with 3-weeks-post-weaning weight in univariable analysis, as well as in the final model. In a previous study conducted by Smith et al,[42] piglets born to primiparous sows had a slower growth rate than piglets from sows with a higher parity. The same was found in the current study, as piglets from higher parity sows had higher 3-weeks-post-weaning body weights than piglets from primiparous sows.

High concentrations of in-feed zinc oxide (≥ 2000 mg per kg) are commonly used therapeutically in starter diets to control post-weaning *Escherichia coli* diarrhea.[43-45] A possible reason why nursery pigs had greater odds of being anemic when consuming high and very high doses of in-feed zinc oxide, compared to a nutritional dose of zinc oxide, is that high doses of zinc interfere with conversion of iron into ferritin.[43] It is also possible that both high and very high levels of zinc in feed may increase the iron requirement in young growing pigs, because zinc decreases the life span of the red blood cell.[43] Zinc, copper, and iron are metals that interact and may present competitive inhibition of transport and bioavailability.[46,47] Therefore, the individual interactions between metals such as copper and zinc may affect iron absorption. In aqueous solutions and at higher doses, competition between metals with similar properties can occur.[48] There are many inhibitory interactions among these metals that could occur when high doses of a certain metal are given.[48] In a competition study, when the concentrations of copper and zinc were increased, iron uptake decreased.[48] In other studies,[49,50] zinc status influenced iron uptake, indicating that DMT1 may not simultaneously transport iron and zinc. A limitation of the current study is that the length of time during which the pigs were fed the nursery diets with the specific concentration of zinc oxide were not measured. The use of high concentrations of in-feed zinc is likely to interfere with iron absorption, and thus should be examined further in future studies.

Copper is another mineral that could have an effect on iron deficiency and anemia in young growing pigs. However, due to the lack of variability of in-feed copper content among the participating farms in this study, copper could not be controlled for. Both copper and zinc are heavy metals that are used therapeutically in feed. Although some herds used nutritional doses of zinc oxide (≤ 500 mg per kg of feed), all herds used high levels of copper sulphate (125 mg per kg of feed).

The economic impact of inadequate iron supplementation in piglets is unknown; however, iron status can be easily evaluated and corrected at minimal additional cost. Moreover, any impact iron deficiency may have on growth rates could negatively affect the cost of production. Therefore, evaluation and correction of iron deficiency or anemia would outweigh the minimal added cost associated with iron supplementation, since this will improve weight gain and overall welfare of the piglets.

In summary, this study identified iron deficiency and anemia in newly weaned pigs and in pigs 3 weeks post weaning. There was evidence that anemia is associated with a negative impact on post-weaning growth performance. The widespread prevalence of iron deficiency and anemia on almost all farms in this study indicates that iron status should be monitored on all farms and supplementation programs assessed.

Implications

- Iron supplementation protocols used by these participating farms were not sufficient to meet iron requirements of large, fast-growing suckling pigs.
- Iron deficiency and anemia may persist beyond 3 weeks in the nursery.
- Anemia is negatively associated with post-weaning growth.
- Under the conditions of this study, high dietary concentrations of zinc oxide (> 2000 mg per kg of feed) are associated with a higher risk of anemia in weaned pigs

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Conflict of interest

None reported

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Original research

Effect of spray-dried porcine plasma protein and egg antibodies in diets for weaned pigs under environmental challenge conditions

David Torrallardona, DVM, PhD; Javier Polo, MSc, PhD

Summary

Objectives: To study the effects on performance of weaned pigs reared in an uncleaned nursery and fed diets containing either egg yolk antibodies (EYA) or spray-dried porcine plasma (SDPP) at one of two dietary inclusion rates.

Material and methods: Weaned pigs (21 days of age; 6.3 kg body weight) housed in an uncleaned nursery were fed diets containing 3% or 6% SDPP or 0.2% EYA for 14 days post weaning, then a common diet to day 28 post weaning (nine replicates, four pigs per pen).

Results: During the initial 14 days, in pigs fed diets with increasing levels of SDPP, there was a linear improvement ($P < .05$) in day 14 body weight and average daily weight gain (ADG) and a tendency ($P < .10$) for improved average daily feed intake (ADFI) and gain-to-feed ratio (G:F). In addition, pigs fed SDPP had greater ADG, ADFI, and G:F than pigs fed EYA ($P < .05$). Performance variables did not differ between pigs fed the EYA diet and those fed the unsupplemented control diet. During the common starter-diet phase (days 15 to 28), G:F was lower ($P < .01$) for pigs previously fed SDPP diets. Over the 28-day period, performance variables did not differ ($P > .05$).

Implications: Under the conditions of this study, while performance may not be better in pigs fed an EYA diet than in pigs fed a control diet, performance may be better in pigs fed SDPP diets than in controls during the initial 14-day period.

Keywords: swine, spray-dried porcine plasma, egg antibodies, environmental stress, post-weaning diets.

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Resumen - Efecto de la proteína de plasma secada por spray y los anticuerpos de huevo en dietas de cerdos destetados bajo condiciones medioambientales de reto

Objetivos: Estudiar los efectos en el desempeño de cerdos destetados criados en un destete sucio y alimentados con dietas conteniendo anticuerpos de yema de huevo (EYA por sus siglas en inglés) o plasma secado por spray (SDPP por sus siglas en inglés) en uno de dos porcentajes de inclusión en la dieta.

Material y métodos: Cerdos destetados (21 días de edad; 6.3 kg de peso corporal) alojados en un destete sucio fueron alimentados con dietas que contenían 3% ó 6% SDPP ó 0.2% EYA durante 14 días post-destete, seguido de una dieta común hasta el día 28 post-destete (nueve réplicas, cuatro cerdos por corral).

Resultados: Durante los primeros 14 días, en cerdos alimentados con dietas con niveles crecientes de SDPP, se observó una mejora linear ($P < .05$) en el peso corporal del día 14 (ADG por sus siglas en inglés), y una tendencia positiva en el consumo diario de alimento (ADFI por sus siglas en inglés) y la relación ganancia-alimento (G:F, por sus siglas en inglés). Además, los cerdos alimentados con SDPP tuvieron mayores ADG, ADFI, y G:F que los cerdos alimentados con EYA ($P < .05$). Las variables del desempeño no difirieron entre los cerdos alimentados con la dieta EYA y los alimentados con la dieta control no suplementada. Durante la fase común de inicio de dieta (días 15 a 28), la G:F fue más baja ($P < .01$) en los cerdos alimentados anteriormente con dietas SDPP. Durante el periodo de 28 días, las variables de desempeño no difirieron ($P > .05$).

Implicaciones: Bajo las condiciones de este estudio, mientras que el desempeño puede no ser mejor en los cerdos alimentados con una dieta con EYA comparado con en los cerdos alimentados con la dieta control, el desempeño durante el periodo inicial de 14 días puede ser mejor en los cerdos alimentados con la dieta SDPP que en los controles.

Résumé - Effet de protéines plasmatiques porcines séchées par jet et d'anticorps d'œufs chez des porcs sevrés dans des conditions environnementales délétères

Objectifs: Étudier les effets sur la performance de porcs sevrés élevés dans une pouponnière non-nettoyée et nourris avec des rations contenant soit des anticorps de jaune d'œuf (EYA) ou un des deux taux de protéines plasmatiques porcines séchées par jet (SDPP).

Matériels et méthodes: Des porcs sevrés (21 jours d'âge; 6,3 kg de poids corporel) logés dans une pouponnière non-nettoyée ont été nourris avec des rations contenant 3% ou 6% de SDPP ou 0,2% de EYA pendant 14 jours post-sevrage, puis avec une ration commune jusqu'au jour 28 post-sevrage (neuf réplications, quatre porcs par enclos).

DT: IRTA, Monogastric Nutrition Subprogram, Mas de Bover, Constantí, Spain.

JP: APC Europe, SA, Granollers, Spain.

Corresponding author: Dr Javier Polo, APC Europe, SA, Avda. Sant Julià 245-258. P. I. El Congost. E-08403. Granollers, Barcelona, Spain; Tel: +34 629 327 346; Fax: +34 93 849 51 83; E-mail: javier.polo@apc-europe.com.

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Résultats: Pendant les 14 premiers jours, chez les porcs nourris avec des rations ayant des quantités de SDPP croissantes, il y avait une amélioration linéaire ($P < 0,05$) du poids corporel au jour 14 et du gain de poids journalier moyen (ADG) et une tendance ($P < 0,10$) à une amélioration de la consommation quotidienne moyenne de nourriture (ADFI) et du ratio gain-consommation (G:F). De plus, les porcs nourris avec SDPP avaient des valeurs d'ADG, d'ADFI, et de G:F plus élevées que les porcs nourris avec EYA ($P < 0,05$). Aucune différence ne fut notée entre les variables de performance des porcs nourris avec la ration avec EYA et ceux nourris avec une ration témoin non-supplémentée. Pendant la période d'alimentation avec la ration de début commune (jours 15 à 28), le ratio G:F était inférieur ($P < 0,01$) pour les porcs nourris avec les rations contenant SDPP. Pour la période entière des 28 jours, les variables de performance ne différaient pas ($P > 0,05$).

Implications: Dans les conditions expérimentales de la présente étude, bien que les performances des porcs nourris avec une ration EYA n'étaient pas meilleures que celles des porcs nourris avec une ration témoin, les performances des porcs nourris avec des rations SDPP peuvent être meilleures que celles des animaux témoins durant la période initiale de 14 jours.

Weaning is one of the most stressful periods in the life of a pig, resulting in lower feed intake, poorer growth, and higher morbidity and mortality, particularly during the first weeks after weaning or until the immune system has become more fully developed. Weaning is a stress, independent of weaning age, caused by the abrupt separation from the sow and by other stressors related to changes in the physical and social environment, mingling with pigs from different litters, dietary transitions, and exposure to different pathogens or antigens.[1] Weaning stress causes intestinal inflammation and damage to mucosal barrier structure and function.[2-5] Therefore, it is crucial that the pig overcomes weaning stress rapidly to survive and be productive through the commercial production cycle.

Along with good husbandry and health management, dietary interventions may be a viable and practical way to help pigs adapt and transition through the complexities associated with weaning stress. Many studies have demonstrated consistently better performance with the use of spray-dried plasma (SDP) than with other protein sources, independent of its origin (porcine, bovine, or a blend), in diets for weaned pigs.[1,6,7] Several studies have also reported that SDP reduces the incidence of diarrhea during the post-weaning phase,[8,9] and improved performance has been described in pigs kept under less hygienic research conditions or commercial circumstances[10] or in younger pigs with less mature immune systems.[11] In addition, some studies also show that the intestinal mucosal immune response is better in animals fed diets containing SDP.[12-14] Taken together, these studies suggest that diets containing plasma proteins improve the immunological response to stress without compromising the response to pathogens, allowing the animal to use more nutrients for growth and other productive functions instead of for maintaining the immune response.

Spray-dried whole egg (SDE) without shells is an excellent nutrient source due to its high digestibility, favorable amino acid balance, fat content, and high metabolizable energy (ME).[15,16] In addition, SDE derived from hens not hyperimmunized against specific pig pathogens contains active components such as immunoglobulins (IgY),[17] lysozyme, and antimicrobial proteins.[18] Inclusion of SDE in nursery pig diets did not consistently promote better performance than did other high-quality protein ingredients, eg, SDP, milk proteins, and fish meal.[19]

Egg-yolk antibodies (EYA) from eggs produced by hens hyperimmunized against specific bacterial antigens have been suggested as a more efficient source of SDE and as an effective way to control diarrhea in post-weaned pigs.[20-23] Positive responses to EYA have been consistent when the microorganism used to challenge the pigs is the same as that used to hyperimmunize the hens.[20-23] However, limited published information is available about the use of EYA under commercial conditions when sanitation is suboptimal.

The purpose of this study was to investigate the effects on performance of weaned pigs reared under unsanitary conditions (pens not cleaned and sanitized after previously housing pigs) and fed diets containing either EYA or one of two dietary inclusion levels of SDP of porcine origin (SDPP).

Materials and methods

The experimental procedures with animals described in this study were conducted after approval from the Institut de Recerca i Tecnologia Agroalimentàries (IRTA) Ethical Committee on Animal Experimentation. The IRTA is a research institute belonging to the Catalonia government.

Animals and housing

The study was conducted in the post-weaning facility of the experimental farm of IRTA Animal Nutrition (Centre Mas de Bover, Constantí, Spain). The pigs were housed in two nursery rooms with 24 and 12 pens, respectively, each 1.7 m^2 (0.96 m × 1.77 m), with four pigs per pen. All pens were identically equipped with one single-sided hopper feeder with four eating spaces, and a cup-type drinker system. The rooms had automatic heating, forced ventilation, and completely slatted floors. The experimental facilities were not cleaned prior to the entry of the study animals in order to impose an environmental challenge.

One hundred and forty-four newly weaned intact male pigs (Duroc × Landrace), obtained from a commercial farm at 21 days of age, were used in the study. Average initial body weight (BW) was 6.3 kg (standard deviation [SD], 0.80 kg). At the start of the study, the pigs were sorted by BW and divided into 36 groups of four animals, so that the first four groups (ie, 16 animals) belonged to block 1, the next four to block 2 and so on, up to block 9. The four animals in a given group were randomly distributed, using the random number generator function in Excel (Microsoft Corporation, Redmond, Washington), among the four replicates (pens) in the corresponding block, and the same was done for the other groups in the same block. This was repeated for all blocks, so that nine blocks with four replicates (pens of four piglets) were generated. The four pens belonging to a block were adjacent to each other and in the same room, so that location was also considered within the block effect. Once the pigs had been assigned to the 36 replicates, the four experimental treatments were randomly assigned, using the random generator function in Excel, to the four replicates in each block.

Experimental design and treatments

The experimental diets (13.8 MJ metabolizable energy [ME]; 13.5 g per kg lysine) were offered for a period of 14 days (pre-starter phase). Between days 15 and 28, a common starter diet (without SDPP or EYA) was offered (13.6 MJ ME;

Table 1: Composition (%) of nursery-pig diets containing spray-dried porcine plasma (SDPP) or egg yolk antibodies (EYA) or an unsupplemented control diet (Control)*

Ingredients	Control	SDPP3	SDPP6	EYA	Starter
Wheat	25.00	25.00	25.00	25.00	0.00
Barley	20.00	20.00	20.00	20.00	38.43
Maize	15.00	15.00	15.00	15.00	25.00
Soybean meal (48% CP)	12.62	13.11	13.25	12.62	22.60
Sweet milk whey	10.00	10.00	10.00	10.00	6.86
Soy protein concentrate (65% CP)	6.00	3.00	0.00	5.80	0.00
SDPP†	0.00	3.00	6.00	0.00	0.00
EYA‡	0.00	0.00	0.00	0.20	0.00
Wheat middlings	4.00	4.00	4.00	4.00	0.00
Lard	3.50	3.50	3.55	3.50	3.39
Dicalcium phosphate	1.86	1.59	1.53	1.86	2.20
Calcium carbonate	0.53	0.71	0.74	0.53	0.18
L-lysine-HCl	0.49	0.40	0.32	0.49	0.40
DL-methionine	0.19	0.15	0.14	0.19	0.15
L-threonine	0.17	0.11	0.06	0.17	0.13
L-tryptophan	0.05	0.03	0.02	0.05	0.01
Salt	0.20	0.00	0.00	0.20	0.24
Vitamin-mineral complex§	0.40	0.40	0.40	0.40	0.40

* 144 weaned pigs (21 days of age; 6.3 kg body weight) were housed in two nursery rooms with 24 and 12 pens, respectively, and four pigs per pen. Pens had not been cleaned after the previous use. Experimental diets were fed during the first 14 days post weaning (pre-starter phase). Between days 15 and 28 post weaning, all animals were fed the Starter diet. Totals may not add to 100.00% because of rounding. Diets were formulated to meet the nutrient requirements of piglets (National Research Council [1998]).[24]

† SDPP used was AP-820P (APC Europe, SA, Granollers, Spain), included as either 3% (SDPP3) or 6% (SDPP6) of the diet.

‡ EYA used was Globigen (EW Nutrition GmbH, Visbek, Germany).

§ Provided per kg of diet: vitamin A, 10,000 IU; vitamin D3, 2000 IU; vitamin E, 15 mg; thiamine, 1.3 mg; riboflavin, 3.5 mg; vitamin B12, 0.025 mg; vitamin B6, 1.5 mg; calcium pantothenate, 10 mg; nicotinic acid, 15 mg; biotin, 0.1 mg; folic acid, 0.6 mg; vitamin K3, 2 mg; iron, 80 mg as iron sulfate; copper, 6 mg as copper sulfate; cobalt, 0.75 mg as cobalt sulfate; zinc, 185 mg as zinc oxide; manganese, 60 mg as manganese sulfate; iodine, 0.75 mg as potassium iodate; selenium, 0.10 mg as sodium selenite; ethoxyquin, 0.15 g.

CP = crude protein.

12.5 g per kg lysine). Diets were formulated to meet the nutrient requirements of the National Research Council (NRC; 1998).[24] Feed was presented in mash form and offered ad libitum. Ingredient and nutritive compositions of the diets are shown in Table 1 and Table 2, respectively.

The four feed treatments consisted of a control group (Control); two treatments containing SDPP (AP820P from APC Europe, SA, Granollers, Spain) at 3% and 6%, replacing soy protein concentrate in the Control diet (SDPP3 and SDPP6 groups); and a fourth treatment containing a commercial EYA product (Globigen; EW Nutrition GmbH, Visbek, Germany) at the manufacturer-recommended dietary inclusion rate (0.2%), also replacing soy protein concentrate (EYA group). According to the manufacturer's literature, the Globigen EYA product contains specific antibodies (immunoglobulins) against a number of pathogens described as "*Escherichia coli K88, E coli K99, E coli 987P, E coli Oedema, Salmonella typhimurium,* transmissible gastroenteritis virus*, Cryptosporidium, Rotavirus, Clostridium perfringens,* circovirus." The experimental feeds were offered from day 1 through day 14 post weaning, and a common starter diet was fed to all animals from day 15 through day 28.

Feed and piglets were weighed at the start of the study, at day 14, and at the end of the study (day 28). Initial and final BW, average daily gain (ADG), average daily feed intake (ADFI), and gain-to-feed ratio (G:F) were calculated.

Statistical analysis

The experimental unit was the pen, and average pen values were used for the performance parameters. For statistical analysis (GLM procedure; SAS Inc, Raleigh, North Carolina), a randomized block design was used, with initial weight and pen location as block criteria. Least squares means, probabilities of differences, and standard errors of the mean were obtained to evaluate differences among treatment means.

In addition, orthogonal contrasts were used to compare production parameters in pigs in the Control or the EYA treatments versus the two SDPP treatments, and to determine

Table 2: Estimated nutritive compositions (%) of an unsupplemented nursery-pig control diet (Control) or the control diet supplemented with either spray-dried porcine plasma (SDPP) or egg yolk antibodies (EYA)*

Analyte	Control	SDPP3	SDPP6	EYA	Starter
Crude protein	19.12	19.34	19.43	19.08	19.50
Crude protein (analyzed)	19.53	19.55	19.62	19.67	18.51
Crude fiber	2.86	2.79	2.70	2.85	3.12
Fat	5.20	5.26	5.35	5.21	5.37
Ash	5.69	5.71	5.98	5.69	5.86
Lactose	7.29	7.29	7.29	7.29	4.32
Energy (MJ ME/kg)	13.8	13.8	13.8	13.8	13.6
Calcium	0.85	0.85	0.85	0.85	0.80
Phosphorous	0.70	0.70	0.74	0.70	0.78
Chloride	0.46	0.41	0.49	0.46	0.41
Sodium	0.15	0.19	0.30	0.15	0.15
Total methionine	0.47	0.43	0.41	0.47	0.43
Total methionine + cystine	0.81	0.81	0.83	0.81	0.75
Total lysine	1.35	1.35	1.35	1.35	1.25
Total tryptophan	0.27	0.27	0.27	0.27	0.23
Total threonine	0.88	0.88	0.88	0.88	0.81
Total valine	0.91	0.96	1.01	0.91	0.88
Total isoleucine	0.80	0.79	0.77	0.80	0.76
SID methionine	0.44	0.40	0.37	0.44	0.40
SID methionine + cystine	0.73	0.71	0.72	0.73	0.61
SID lysine	1.23	1.21	1.19	1.23	1.12
SID tryptophan	0.24	0.23	0.22	0.24	0.20
SID threonine	0.76	0.74	0.73	0.76	0.70
SID valine	0.79	0.83	0.86	0.79	0.72
SID isoleucine	0.71	0.69	0.67	0.71	0.65

* Study described in Table 1. Diets were formulated to meet the nutrient requirements of piglets (National Research Council [1998]).[24]
SID = standardized ileal digestible amino acids; ME = metabolizable energy.

the linear response to the increasing dose of SDPP supplementation. Results were considered statistically significant at $P < .05$ and a trend was defined at $P < .10$.

Results

During the pre-starter period (days 0 to14), four piglets died with signs of diarrhea (two in the Control group and two in the SDPP3 group). During the common starter phase (days 15 to 28), another four piglets died (one in the Control group, two in the SDPP6 group, and one in the EYA group). Four animals from the SDPP3 treatment group were culled due to extremely poor body condition. The data from these animals were considered missing values and were not used in the calculations. Their feed intake was calculated with a model that estimates the individual feed intake of pigs in group feeding,[25] and was subtracted from the total intake of the corresponding pen.

Between days 0 and 14 (Table 3), ADG was greater in the SDPP6 group ($P < .05$) than in the Control and EYA groups. For BW, ADG, and ADFI, performance in the SDPP3 group was intermediate between the Control and SDPP6 groups. Gain-to-feed ratio for EYA treatment was the lowest in this period. Linear improvements ($P < .05$) in BW and ADG and tendencies ($P < .10$) for higher ADFI and G:F were observed with increasing SDPP during this period. In addition, groups fed SDPP had greater ADG, ADFI, and G:F than the group fed EYA ($P < .05$), and tended to have greater ADG than the Control group ($P < .10$). Performance data did not differ between pigs fed the diet including EYA and those fed the Control diet.

During days 15 to 28 (Table 4), when a common starter diet was fed to all animals, no significant differences in ADG and ADFI were observed among treatments. However, pigs that had been previously fed SDPP had lower G:F than did the Control and EYA groups ($P < .01$).

For days 0 through 28, no differences for any parameters were observed among treatment groups. However, relative to the Control

group, in pigs previously fed SDPP6, final BW was 0.46 kg higher, while in pigs previously fed the SDPP3 and EYA diets, final BW was 0.59 and 0.43 kg lower, respectively.

Discussion

In this study, pigs were subjected to challenging conditions by being weaned and housed in pens uncleaned since previous use. These conditions were effective in inducing post-weaning growth depression, as can be observed from the generally low growth rates of these animals, compared to normal production values in the industry or experience before and after the current trial in the same facilities. In the study facility, ADG of similar pigs on similar diets, but weaned into a clean environment, ranged from 175 to 250 grams per day at 0 to 14 days post weaning. The pathogens associated with these stressful conditions were not determined, but signs of watery diarrhea and poor feed conversion were observed.

Although the nutrient compositions of the experimental diets met the nutrient recommendations of the NRC (1998),[24] it could be argued that the diets were limiting for sodium and some essential amino acids, according to the values proposed more recently (NRC; 2012).[26] Even if some nutrients were limiting, the magnitude of their limitation cannot explain the poor performances observed. Instead, it is more likely that poor performance was driven mainly by the markedly lower feed intake of the study pigs. Under the conditions of the current trial, health status of the animals might have been compromised, resulting in the observed poor appetite.

The better performance observed with the inclusion of SDPP in the feed during the initial 14 days was consistent with previous publications indicating that formulating diets with SDPP improves post-weaning performance of pigs, especially when sanitary conditions are not optimal.[1,10] Several publications have demonstrated that when SDPP is included in the diet of animals challenged with a diversity of pathogens (*Escherichia coli*, *Salmonella*, rotavirus, porcine reproductive and respiratory syndrome, and porcine epidemic diarrhea virus [PEDV]), the animals had better health and more rapid recovery from these pathogens.[27-33] Enhanced performance provided by SDPP in diets for weaned pigs may be related to beneficial effects on intestinal barrier function, inflammation, and diarrhea.[13]

During the common starter phase (days 15 through 28), the two groups of pigs previously fed SDPP diets had lower G:F than did the Control or EYA groups. Over the entire study period (days 0 to 28), pigs previously fed the SDPP6 diet had numerically better final BW, ADG, and ADFI than did the Control group, but this was not the case for the SDPP3 and EYA groups. The reasons for these observations in the current study are unknown. However, it has been reported[13] that pigs fed a diet with 5% SDPP for 14 days post weaning had less secretory activity, lower diarrhea scores, and less pro-inflammatory cytokine (mRNA TNF-α) in colon tissue, resulting in less damage to gut barrier function than in pigs fed diets with either 0% or 2.5% SDPP.[13] In addition, a recent study[30] demonstrated that the dietary inclusion rate of SDPP and feeding duration post weaning is important for maintaining longer-term gastrointestinal tract function after SDPP is removed from the diet. In that study,[30] pigs were fed diets with either 0%, 2.5%, or 5.0% SDPP (5.0% SDPP in a diet fed for 14 days versus 2.5% SDPP in a diet fed for 7 days), and at day 34 in the nursery (50 days of age) pigs were transported to a different facility and challenged with *Salmonella* Typhimurium. At day 2 post challenge, distal ileum samples were collected and subjected to various chemical and physical measures. Results indicated pigs previously fed the 5.0% SDP diet for 14 days post weaning had lower histological scores, myeloperoxidase and IL-8 concentrations, and 4 kDa fluorescein isothiocyanate dextran (FD4) flux rates, along with higher concentrations of plasma and ileal TNF-α than in other groups, suggesting that inclusion rate and duration of feeding SDPP in diets can influence subsequent immunological and intestinal injury induced by *Salmonella* challenge. The data from the

Table 3: Productive parameters (least squares means) of pigs between 0 and 14 days of the experiment*

	BW (kg)		ADG (g)	ADFI (g)	G:F ratio
	Day 0	Day 14			
Control	6.31	6.61[a]	21[a]	108	0.26
SDPP3	6.31	6.81[ab]	35[ab]	123	0.28
SDPP6	6.33	7.19[b]	62[b]	141	0.43
EYA	6.30	6.44[a]	9[a]	98	0.18
Root MSE	0.031	0.496	34.1	36.8	0.166
			***P* values**		
Treatment effect	> .10	< .05	< .05	< .10	< .10
Linear effect of SDPP dose	> .10	< .05	< .05	< .10	< .10
SDPP versus Control	> .10	< .10	< .10	> .10	> .10
SDPP versus EYA	> .10	< .05	< .05	< .05	< .05

* Study described in Table 1. Data were analyzed as a randomized block design with pen as the experimental unit using the GLM procedure (SAS Institute Inc, Cary, North Carolina). Values were considered significant at $P < .05$, and $P < .10$ was considered a trend.

[ab] Values in the same column with different superscript letters are significantly different ($P < .05$).

BW = body weight; ADG = average daily gain; ADFI = average daily feed intake; G:F = gain to feed ratio; SDPP = spray-dried porcine plasma, at 3% (SDPP3) or 6% (SDPP6) of the diet; EYA = egg yolk antibodies; MSE = mean square error.

Table 4: Productive parameters (least squares means) of pigs between 15 and 28 days of experiment*

	BW (kg)		ADG (g)	ADFI (g)	G:F ratio
	Day 15	Day 28			
Control	6.61[a]	9.82	229	357	0.64[a]
SDPP3	6.81[ab]	9.23	173	330	0.53[b]
SDPP6	7.19[b]	10.28	221	392	0.55[b]
EYA	6.44[a]	9.39	211	336	0.63[a]
Root MSE	0.496	1.151	53.5	76.8	0.060
			P value		
Treatment effect	< .05	> .10	> .10	> .10	< .01
Linear effect of SDPP dose	< .05	> .10	> .10	> .10	< .01
SDPP versus Control	< .10	> .10	> .10	> .10	< .01
SDPP versus EYA	< .05	> .10	> .10	> .10	< .01

* Study described in Table 1. Data analyzed as a randomized block design with pen as the experimental unit using the GLM procedure (SAS Institute Inc, Cary, North Carolina). Values were considered significant at $P < .05$, and $P < .10$ was considered a trend.

[ab] Values in the same column with different letters are significantly different ($P < .05$).

BW = body weight; ADG = average daily gain; ADFI = average daily feed intake; G:F = gain to feed ratio; SDPP = spray-dried porcine plasma, at 3% (SDPP3) or 6% (SDPP6) of the diet; EYA = egg yolk antibodies; MSE = mean square error.

current study indicate that the inclusion rate of SDPP in the diet may need to be higher than 3% to maintain the performance benefits obtained during the initial 14 days post weaning.

Likewise, some studies suggest a growth-promoting effect of EYA in early-weaned pigs challenged with specific pathogens.[20-23] In most studies demonstrating improvements in animal performance when EYA has been fed, the animals had been challenged with the same pathogen for which the hens had been inoculated to produce the specific EYA. For example, a study[22] reported the effect on performance of pigs challenged with enterotoxigenic *Escherichia coli* (ETEC) K88 when a pea-protein-based diet was supplemented with EYA from hens immunized with ETEC K88 antigen. The results indicated that the pigs fed a diet with EYA had higher ADG than those fed a Control diet without EYA. In the same study,[22] pigs fed a diet with SDPP also had a higher ADG than the Control group, but no differences were observed between the groups supplemented with SDPP or EYA. In addition, the authors reported less severe diarrhea and lower mortality when either EYA or SDPP was included in the diets. Both the EYA and SDPP diets contained specific antibodies against ETEC K88 and F18, and therefore the authors suggested that the antibodies in these two products may have prevented ETEC K88 from binding to the mucosal receptors.[22] A different study[19] in weaned pigs fed diets containing SDE without antibodies against specific pathogens reported that pigs fed the SDE diet had higher ADFI during the first 7 days post weaning than did the pigs fed the control diet, but other performance parameters did not differ. However, pigs fed a diet containing SDP had significantly higher ADG, ADFI, and G:F than the control group. Similarly, in an earlier study,[34] weaned pigs were fed a diet containing egg-yolk powder with antibodies against *Salmonella* Typhimurium or a diet containing SDP, or were treated with antibiotics starting at day 3 of the trial. At day 7, all pigs were challenged with the strain of *Salmonella* Typhimurium used in the egg-yolk powder. The authors found that the percentage of pigs shedding *Salmonella* was lower in the antibiotic treatment group than in the other groups. However, *E coli* antibiotic resistance was greater in pigs fed antibiotics than in pigs in the other treatment groups. Health and performance indicators (weight gains, white blood cell counts, and plasma concentrations of *Salmonella* antibodies) did not differ among treatment groups, indicating that feeding those antibodies may not have been effective in reducing *Salmonella* shedding. Further, in another study,[35] weaned pigs challenged with $K88^+$ *E coli* and fed diets with 0.00%, 0.32%, or 3.20% EYA (IgY) developed watery diarrhea and became dehydrated, compared to an unchallenged Control group. No quantifiable concentrations of IgY were detected either in treated or Control pigs by testing small intestinal content using an enzyme-linked immunosorbent assay. According to the authors, the *E coli* challenge was successful in creating a clinical syndrome similar to field cases. The presence of chicken egg-yolk antibodies in the feed did not appear to be effective in preventing the disease.

During the process of manufacturing commercial spray-dried plasma, one single production lot of SDPP is derived from the pooled blood of 6000 to 10,000 pigs.[36] Each lot of SDPP contains antibodies against multiple pathogens circulating in the pig population at any time. Therefore, it is not surprising that SDPP may contain antibodies with neutralizing capacity against the unknown pathogens affecting the pigs used in our study, as it has been demonstrated that commercial SDPP contains neutralizing antibodies against common swine pathogens.[37] However, there is substantial literature demonstrating that the effects of dietary SDPP are due to the presence not only of immunoglobulins, but also other functional proteins, growth factors, cytokines, and biologically active compounds like functional peptides and amino acids

that contribute to its beneficial effects on animal performance and health.[12,38] However, the exact roles of each of the functional components present in plasma that contribute to the physiological improvements of gastrointestinal barrier function have yet to be completely elucidated. These proteins can interact with immune cells in the mucosa, thus changing the cytokine environment. In addition to this luminal effect, spray-dried plasma also has systemic effects. For example, it can reduce the expression of pro-inflammatory cytokines in peripheral tissues of pigs challenged with lipopolysaccharides (LPS) from *E coli*[39] and prevent the increase in activated lymphocyte populations in an LPS-induced lung inflammation model.[40] In addition, it has been demonstrated that dietary SDP decreased the uterine concentrations of TNF-α and IFN-γ, and serum TNF-α, C-reactive protein, and cortisol, but increased uterine anti-inflammatory cytokine (TGF-β1) concentration in a pregnancy animal model study. [41] Since the organized gut-associated lymphoid tissue is an inductive site that connects with both local and peripheral effector sites (respiratory tract, glandular tissues, and the uterine mucosa), it can be further hypothesized that spray-dried plasma may favorably modulate the broader common mucosal immune system.

A recent study[33] evaluated a potential positive effect of SDPP or EYA on pigs challenged with PEDV. That EYA product consisted of a liquid egg formulation that, according to company specifications, was tested to contain anti-coronavirus antibodies. The pigs in all the infected groups (Control, SDPP, and EYA) began shedding PEDV in feces by day 3 post infection, and under the study conditions, SDPP or EYA addition did not significantly alter PEDV shedding or overall disease course after experimental challenge, except that the pigs in the SDPP group appeared more active during the acute PEDV disease stage, with less pronounced diarrhea. In addition, fecal PEDV shedding in treated pigs (SDPP or EYA) was lower than in the Control pigs in the early stage of infection, which could contribute to lower environmental PEDV loads and lower transmission rates to uninfected contact pigs. Furthermore, in the same study,[33] at day 3 post infection, adherent *E coli* in the intestine were detected in two of three pigs fed the Control diet and two of three pigs in the EYA group, but none (zero of three) in the SDPP-fed group, indicating that in the PEDV-challenged pigs fed the diet with SDPP, concurrent opportunistic pathogens like *E coli* were prevented.

In summary, the results of the current study indicate that, under the unsanitary conditions described, performance of pigs fed diets with SDPP during the initial 14 days after weaning was better than that of the EYA and Control groups, but longer-term performance (days 0 to 28) did not differ among groups. In this study, challenge pathogens were not identified and antibodies against those specific pathogens were not identified in the EYA fed. Under these conditions, better pig performance was not observed when diets were supplemented with EYA.

Implications

- Under the conditions of this study, spray-dried porcine plasma linearly increases performance in weaned pigs reared in an unknown challenge environment during the period that the ingredient is fed.
- Under the conditions of this study, the performance benefit of feeding spray-dried porcine plasma from day 0 to 14 post weaning is maintained to day 28 when the inclusion rate is 6%, but not when the inclusion rate is 3%.
- Under the conditions of this study, egg-yolk antibodies from eggs of hens hyperimmunized with specific bacterial antigens do not benefit performance in a non-specific pathogenic environment.

Conflict of interest

Javier Polo is employed by APC Europe, SA, Granollers, Spain, a company that manufactures and sells spray-dried plasma. However, APC Europe did not have any additional role in the study design, data collection and analysis, decision to publish, or preparation of the manuscript. David Torrallardona has no conflict of interest.

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Peer reviewed

Case study

Evaluation of responses to both oral and parenteral immunization modalities for porcine epidemic diarrhea virus in production units

Gail Scherba, DVM, PhD; Corinne R. Bromfield, DVM; Vickie L. Jarrell, PhD; Clifford F. Shipley, DVM, Diplomate ACT

Summary

The immune responses (serum anti-porcine epidemic diarrhea virus [PEDV] immunoglobulin G [IgG] and milk antiviral neutralizing antibodies) induced by various combinations of two PEDV immunization modalities (vaccine and oral immunization) were examined in unrelated swine production units in different locations. Anti-PEDV antibodies were undetectable in serum and milk of the control group (non-vaccinated and non-infected). Sows in the unit that received only the PED vaccine (iPED+; Harrisvaccines, Inc, Ames, Iowa) (two doses) remained naive for the wild-type virus and did not develop milk anti-PEDV neutralizing immunoglobulin titers as high as those in the other three production units, which had received oral immunization. Milk anti-PEDV antibody titers in the orally immunized sows appeared to be of longer duration than serum antiviral IgG concentrations. This indicates that oral immunization may be the more efficacious PEDV immunization modality, especially with regard to the production of milk antiviral antibody levels.

Keywords: swine, porcine epidemic diarrhea virus, immunofluorescent antibody assay, oral immunization, parenteral vaccine

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Resumen - Evaluación de la respuesta a las dos modalidades de inmunización parenteral y oral contra el virus de la diarrea epidémica porcina en unidades de producción

La respuesta inmune (suero contra el virus de la diarrea epidémica porcina [PEDV por sus siglas en inglés], inmunoglobulina G [IgG por sus siglas en inglés], y anticuerpos neutralizantes antivirales en la leche) inducida por varias combinaciones de dos modalidades de inmunización del PEDV (vacuna e inmunización oral) fueron examinadas en unidades de producción porcina no relacionadas, y en diferentes ubicaciones. Los anticuerpos contra el PEDV no se detectaron en suero y leche en el grupo control (no vacunados y no infectados). Las hembras en la unidad que recibieron únicamente la vacuna contra PED (iPED+; Harrisvaccines, Inc, Ames, Iowa) (dos dosis) y que no tuvieron contacto con el virus de campo no desarrollaron en leche, una carga de Igs neutralizantes contra el PEDV tan alta, al compararlas con las de las hembras, en las otras tres unidades de producción, que recibieron inmunización oral. Las cargas de anticuerpos contra el PEDV de leche en las hembras inmunizadas oralmente parecen ser de más larga duración que las concentraciones de IgG antivirales en suero. Esto indica que la inmunización oral puede ser la modalidad de inmunización PEDV más eficaz, especialmente en lo que se refiere a la producción de los niveles de anticuerpos antivirales de la leche.

Résumé - Évaluation des réponses aux modalités d'immunisation orale et parentérale contre le virus de la diarrhée épidémique porcine dans des unités de production

Les réponses immunitaires (immunoglobulines G [IgG] sériques anti-virus de la diarrhée épidémique porcine [VDEP] et anticorps neutralisants antiviraux du lait) induites par diverses combinaisons de deux modalités d'immunisation contre le VDEP (vaccin et immunisation orale) ont été examinées dans des unités de production porcine non-apparentées dans des localisations différentes. Les anticorps anti-VDEP étaient non-détectables dans le sérum et le lait des animaux témoins (non-vaccinés et non-infectés). Les truies dans les unités qui ont reçu uniquement le vaccin DEP (iPED+; Harrisvaccines, Inc, Ames, Iowa) (deux doses) et qui sont demeurées naives pour la souche sauvage du virus, ne développèrent pas de titres d'Ig anti-VDEP neutralisants dans le lait aussi élevés que celles dans les trois autres unités de production qui avaient reçu une immunisation orale. Les titres d'anticorps anti-VDEP dans le lait des truies immunisées par voie orale ont semblé durer plus longtemps que les concentrations

GS: Veterinary Diagnostic Laboratory, College of Veterinary Medicine, University of Illinois, Urbana, Illinois.

GS: Department of Pathobiology, College of Veterinary Medicine, University of Illinois, Urbana, Illinois.

CRB, CFS: Agricultural Animal Care and Use Program, University of Illinois, Urbana, Illinois.

VLJ: Department of Animal Sciences, College of Agricultural, Consumer and Environmental Sciences, University of Illinois, Urbana, Illinois.

Corresponding author: Dr Gail Scherba, Department of Pathobiology, College of Veterinary Medicine, University of Illinois, 2001 South Lincoln Avenue, Urbana, IL 61802; Tel: 217-244-0929; Fax: 217-244-7421; E-mail: scherba@illinois.edu.

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d'IgG antivirales sériques. Ceci indique que l'immunisation orale pourrait être la modalité la plus efficace d'immunisation contre le VDEP, surtout en ce qui concerne la production d'anticorps antiviraux dans le lait.

Since the incursion of porcine epidemic diarrhea virus (PEDV) into the United States in May 2013,[1] its clinical disease presentation and pathology have appeared indistinguishable from those of another coronavirus, transmissible gastroenteritis virus (TGEV).[1,2] Both cause significant enteric disease in the young animal, with 30% to 100% mortality in newborn and early-weaned pigs in naive herds.[1] Although both viruses are classified in the *Alphacoronavirus* genus, they are antigenically distinct.[1] In addition, empirical observations from swine practitioners and researchers indicate that a protective immune response to PEDV, unlike the response to TGEV, is of short duration. Animals that have recovered from an infection may be re-infected and manifest clinical disease just months later. Nevertheless, viral family characteristics suggest that immune responses and disease prevention approaches similar to those used against TGEV may be successful.

Coronaviruses infect their hosts by the oral route, with direct invasion of enterocytes from the intestinal lumen, and hence do not require viremic systemic spread. Transmission occurs through virus shedding in the feces where these enveloped viruses can remain highly infectious. Such pathogenesis suggests that mucosal immunity (secretory immunoglobulin A [IgA]) would be important, as opposed to serum antibodies (IgG). For example, previous studies have shown that serum antibodies do not provide significant protection against TGEV infections.[3,4] Consequently, initial protection of the neonate depends upon receiving colostrum-derived neutralizing anti-viral antibodies. The colostrum and milk of sows orally inoculated or naturally infected with virulent TGEV contains primarily secretory IgA (considered optimal lactogenic immunity due to the resistance of IgA to proteolytic degradation in the neonatal gut), whereas colostrum and milk of sows that receive parenteral TGEV inoculation contains mainly IgG antibodies that do not persist in high levels.[3]

Traditionally, TGEV outbreaks in production units were successfully controlled by oral immunization of gilts and sows through feeding intestinal tracts from euthanized infected neonates.[1] This method stimulated mucosal (gut-associated lymphoid tissues [GALT]) immunity in the dam.[5] Such production of colostral anti-TGEV IgA and IgG antibodies would help protect the neonates from clinical disease. Consequently, this strategy has been employed to control PEDV outbreaks. However, advancements in vaccinology have presented a new approach. Specifically, Harrisvaccines, Inc, Ames, Iowa, generated a PEDV vaccine (iPED+) consisting of a transcriptional unit of the viral S (spike) gene encapsulated into particles for parenteral administration to gilts and sows. The PEDV S gene was selected because it encodes the viral attachment protein, a major neutralization target of the immune response. Although the company has received a conditional marketing license for this first-generation vaccine from the US Department of Agriculture, published field data are lacking. The parenteral route of immunization suggests that mucosal immunity may not be sufficiently engaged in the vaccinated animals for their disease protection, yet may produce adequate anti-PEDV antibody titers in colostrum or milk or both for protection of their neonates.

This case study examined the immune responses of sows in four independent production units to the various combinations of two immunization modalities (parenteral vaccine or oral immunization). Anti-PEDV antibody titers were determined in both serum and milk samples to elucidate whether oral immunization with infectious PEDV is required to develop detectable milk neutralizing-antibody titers or if parenteral iPED+ vaccination alone is sufficient.

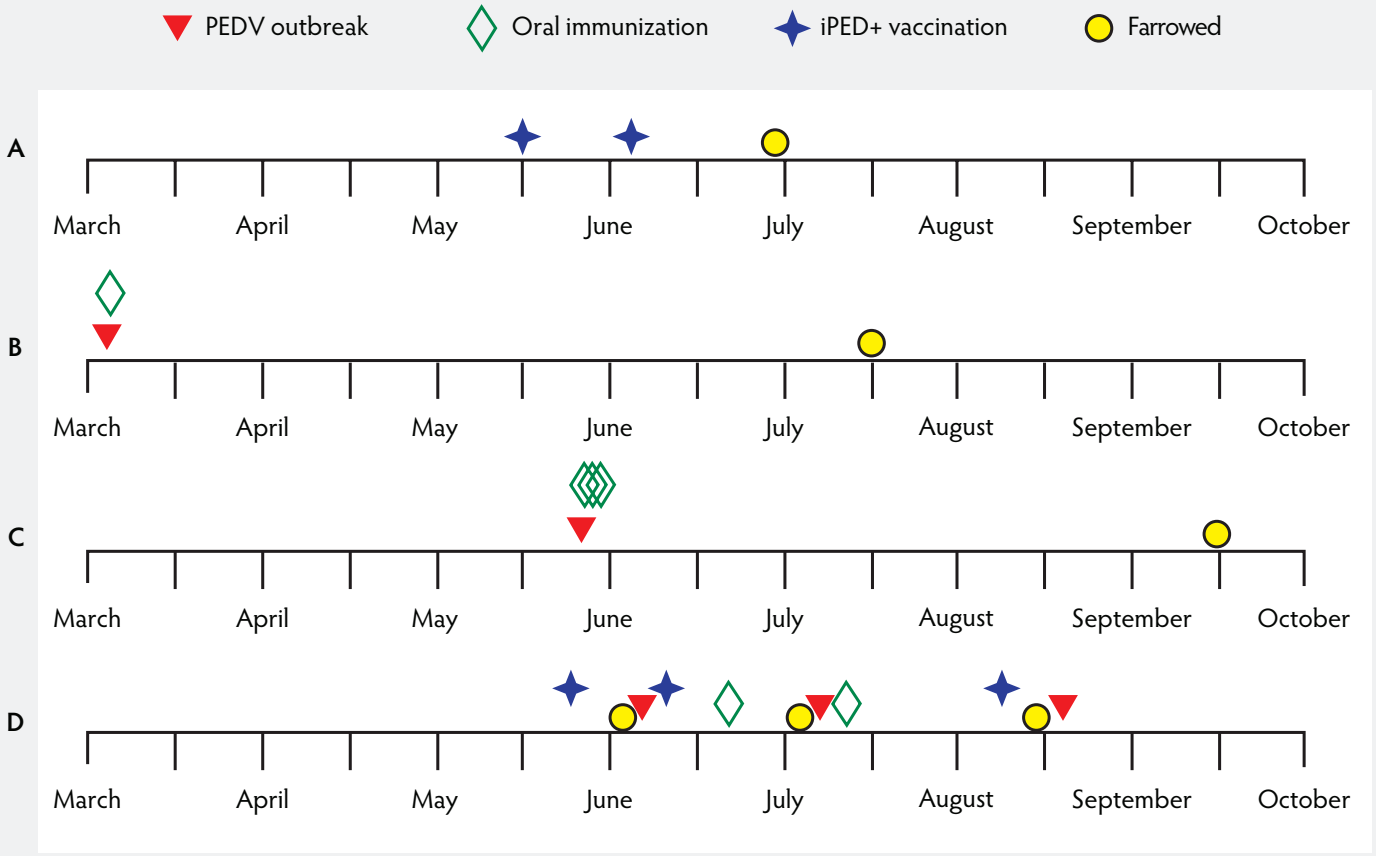
Case farm systems

This project was reviewed and approved by the Institutional Animal Care and Use Committee of the University of Illinois.

Four swine production units were investigated. During the investigation period, Unit A remained naive (non-infected) to the wild-type virus, whereas the other three units (units B, C, and D) had experienced recent PEDV outbreaks. These four units provided a significant opportunity to evaluate the immune responses of sows following various combinations of the two immunization modalities (parenteral first generation iPED+ vaccine and oral immunization using neonatal intestines or contents). Since sampling was conducted in active production units, as opposed to experimentally designed cohorts, treatment group sizes varied; however, each group included at least six animals. A control group, consisting of gilts from an isolated research facility (n = 3) that were neither infected nor vaccinated, served to provide baseline data. Samples included serum (collected 48 hours and 3 weeks post partum) and milk (collected 48 hours post farrowing) from the sows in this study. We selected 48-hour samples so as not to interfere with colostral intake by the neonates, as colostrum begins to be replaced by milk approximately 24 to 36 hours post partum.[6] Fecal swab samples from sows to monitor PEDV fecal shedding were collected only for unit D.

Unit A was a closed-herd, farrow-to-finish production unit of approximately 150 breeding females. The unit maintained three full-time dedicated workers (did not work at other farms). Feed was purchased from a central feed mill. Semen was purchased from boar stud farms free of porcine reproductive and respiratory syndrome virus (PRRSV) and PEDV. Most sows were parity 2 to 4; sows were rarely retained past parity 6. The unit farrowed approximately 25 to 30 sows or gilts every 5 weeks, annually adding approximately 60 replacement females. Gilts were vaccinated with FarrowSure Gold B (Zoetis, Inc, Kalamazoo, Michigan) 5 and 2 weeks prior to breeding to protect against parvovirus, erysipelas, and six *Leptospira* serovars, including bratislava. Gilts were also vaccinated with Litter Guard LTC (Zoetis) at 5 and 2 weeks pre-farrowing to provide protection for the piglets against *Escherichia coli* and *Clostridium perfringens* Type C. Sows received boosters with the same vaccines for each subsequent gestation. Both gilts and sows were boostered with Toxivac AD and E (Boehringer Ingelheim Vetmedica, Ames, Iowa) before each gestation to protect against *Bordetella bronchiseptica*, *Erysipelothrix rhusiopathiae*, and *Pastueurella multocida*. Neonates were vaccinated with CircoFlex (Boehringer Ingelheim Vetmedica) to protect against circovirus and Toxivac AD and E prior to weaning. The sows and gilts, six of which were included in this study, received two doses of first-generation iPED+ vaccine 3 weeks apart (per manufacturer's recommendation), at 6 and 3 weeks pre-farrowing (Figure 1A). Milk and serum samples were collected 48 hours post partum, and serum samples were again collected 3 weeks later.

Figure 1: Timeline of events for the four production units in a case study to determine whether oral immunization of gilts or sows with infectious porcine epidemic diarrhea virus (PEDV) is required to develop detectable milk neutralizing-antibody titers, or if parenteral iPED+ vaccination (Harrisvaccines, Inc, Ames, Iowa) alone is sufficient. The approximate temporal occurrences of one or more natural PEDV outbreaks, oral immunization by feeding intestinal tracts from euthanized PEDV-infected neonates, parenteral administration of first generation iPED+ vaccine, and farrowing within each production unit (A, B, C, and D) are shown.



Unit B was a closed-herd, farrow-to-finish production system consisting of animals that were not vaccinated against PEDV prior to experiencing their first PEDV outbreak in March 2014; disease status was determined by diagnostic testing. The unit lacked any connection with the other production units. The farm internally generated their replacement gilts and used artificial insemination (AI) (semen purchased from boar stud farms free of PRRSV and PEDV), with the exception of natural service in an outdoor breeding system. Each month the unit bred approximately 30 gilts and batch-farrowed 150 sows or gilts or both, for an annual target of approximately 2000 liters. Gilts were vaccinated with PRRS MLV (Ingelvac), FarrowSure Gold B, autogenous swine influenza A virus, and CircoFlex at the time of selection and 1 month later. Sows were vaccinated with FarrowSure Gold B prior to breeding. All breeding animals were vaccinated quarterly with PRRS MLV. Both gilts and sows were vaccinated against swine influenza A virus and with ProSystem (Merck, Kenilworth, New Jersey) and Porcine Ecolizer 3 (Novartis, Greenfield, Indiana) at 5 and 2 weeks pre-farrowing. The PEDV outbreak initially involved second parity or greater sows, but eventually affected all parities, and occurred approximately 4 months pre-farrowing for the six sows included in this study. One oral immunization was performed in the herd immediately after the March 2014 outbreak by feeding back the intestines from clinically affected, euthanized neonates (Figure 1B); PEDV real-time reverse transcriptase-polymerase chain reaction (rRT-PCR) cycle threshold (Ct) values were not determined. Decontamination protocol consisted of routine disinfection of all barns after the outbreak. Serum and milk samples were obtained from the sows 48 hours post partum.

Unit C was a 2650-head, breed-to-wean sow farm that was one of two in the system. Standard biosecurity practices consisted of dedicated caregivers, showers, washing trailers, and composting on site. The unit milled feed on-site that fed this farm as well as its off-site wean-to-market barns. The animals ranged from parity 1 (post wean) through 8, with all breeding done by AI (semen purchased from boar stud farms free of PRRSV and PEDV). The herd had no gilts on site, rather, on a weekly basis, receiving 26 first-parity sows from a separate, PEDV-free sow herd (parity segregation). Prior to receipt, gilts were vaccinated at 15 and 18 weeks of age with PRRS MLV (Ingelvac), at 20 weeks of age with FarrowSure Gold and iFluVent (Harrisvaccine, Ames, Iowa), at 22 weeks of age with Circumvent PCVM G2 (Merck), and at 24 weeks of age with FarrowSure Gold and iFluVent. Sows received ProSystem CE (Merck) and iFluVent at 5 and 4 weeks pre-farrowing, respectively. Annually (September), the herd was boostered with PRRS MLV. Even though the farm was

PEDV-negative, management also included routine feedback, which included feeding mummies and feces to 22-week old gilts prior to breeding, and feeding scour material from neonates to sows at 6, 5, and 4 weeks pre-farrowing. The animals were not vaccinated against PEDV prior to experiencing their first PEDV outbreak in late May 2014. Disease status was determined by diagnostic testing. Sows were orally immunized on each of the 3 days after the outbreak began, approximately 12 weeks pre-farrowing for the six sows included in this study (Figure 1C); PEDV rRT-PCR Ct values were not determined. This PEDV immunization protocol was then discontinued as the farm worked to eradicate the virus. In addition, all neonates were euthanized for 14 consecutive days after the outbreak. Moreover, farrowing and gestation barns were aggressively washed and disinfected during this 12-week down period. Serum and milk samples were obtained from the sows 48 hours post partum.

Unit D was a closed-herd, farrow-to-finish production system of approximately 40 breeding females. The unit maintained two full-time, dedicated workers. Feed was purchased from a central feed mill. Semen was purchased from boar stud farms free of PRRSV and PEDV. Most sows were parity 2. The unit farrowed approximately 8 to 12 sows or gilts or both every 6 weeks, annually adding approximately 20 replacement females. Gilts were vaccinated with FarrowSure Gold 5 and 2 weeks prior to breeding. For each successive breeding, animals were boostered with Litter Guard LTC at 5 and 2 weeks pre-farrowing. Before each gestation, sows were boostered with the same vaccines, and both gilts and sows were boostered with Toxivac AD and E. Piglets were vaccinated with CircoFLEX prior to weaning and then vaccinated with Parasail (Newport Laboratories, Worthington, Minnesota) to protect against *Hemophilus parasuis* in the nursery. Each pregnant sow and gilt was vaccinated with the first-generation iPED+ vaccine. Within 2.5 weeks after their first iPED+ immunization, sows in the farrowing unit became PEDV-infected through a natural outbreak (June 2014), during which both adults and neonates manifested severe acute PEDV disease (Figure 1D). Initially the infection remained limited to the farrowing unit, but all pigs on the farm were exposed intentionally at this time (included gestation, growers, and finishing units). All neonates were euthanized and their intestinal contents were harvested. The infection status of the neonates born during this outbreak was verified by histopathology, bacterial culture, and PEDV rRT-PCR testing as routinely performed at the University of Illinois Veterinary Diagnostic Laboratory (UI VDL). Each pregnant sow in the gestation unit received 10 mL of intestinal slurry (Ct value of 18) added to their individual feed. Also, the farrowing unit was decontaminated prior to entry of the next group of sows. Subsequently, a second outbreak (July) occurred 4 weeks after the initial wild-type infection and 2 weeks after oral immunization of all gestating sows, including the next group of sows that farrowed just prior to this outbreak. This time, clinical disease was limited to the neonates in the farrowing unit; however, they manifested much milder clinical disease than that observed in the first outbreak. As before, all neonates were euthanized, then the farrowing unit was decontaminated using the same procedure used after the first outbreak. All sows in the gestation unit received their second oral immunization with intestinal contents, as described, 5 weeks before the next farrowing. In addition, they received a third iPED+ vaccination approximately 1 week before the third group of sows farrowed. Therefore, the pregnant sows in this third group (nine of which were included in this study) had received prepartum three doses of iPED+ and two oral immunizations. Serum and fecal-swab samples were obtained 1 day before oral immunization (week 0) and weekly thereafter for a total of five sampling time points, with the last (4 weeks) just prior to farrowing and the third PEDV outbreak (August; Figure 1D). Porcine epidemic diarrhea virus infections during the outbreaks were identified only by molecular assay; none of the S genes were sequenced. Milk samples were obtained 48 hours post partum. Decontamination efforts after each outbreak included power-spray washing using 1-Stroke Environ (Steris Corp, St Louis, Missouri) to wash walls and floors. Additionally, after the second and third outbreaks, washing was followed by 160°C heat treatment of each farrowing room. Environmental samples for molecular PEDV testing were collected from farrowing, gestation, nursery, and office, and other traffic areas after the second and third clean-up efforts.

Laboratory assays

The humoral (IgG) immune response to PEDV was quantified by using an immunofluorescent antibody assay (IFA; VMRD, Inc, Pullman, Washington) performed at the UI VDL. Similar to ELISA systems, such assays detect any anti-virus antibodies, therefore neutralizing antibody levels cannot be specifically quantified. Serum samples were tested in duplicate using twofold dilutions from 1:40 to 1:320. Samples that lacked a detectable antibody response at the 1:40 dilution were tested at a 1:20 dilution. Samples were considered negative if anti-PEDV IgG antibodies were undetectable at the 1:20 dilution.

Maternal immunoglobulin (IgG, IgA, IgM) response to PEDV was also evaluated in milk samples obtained from sows 48 hours post parturition. Antibodies to PEDV were measured using a PEDV fluorescent focus neutralizing (FFN) assay performed at the South Dakota State University Animal Disease Research and Diagnostic Laboratory. Such assays detect neutralizing anti-virus antibodies. Samples were considered negative if anti-PEDV neutralizing antibodies were undetectable at a 1:40 dilution.

A PEDV rRT-PCR assay routinely performed at the UI VDL was used to assess virus shedding in fecal samples. Samples were considered positive for detection of the viral genome if Ct values were ≤ 37 and negative if Ct values were > 40. Counts of 38 and 39 were considered suspect, and retesting may be suggested.

Statistical analysis was not performed, given the type of data obtained from this case study in active production units.

Antibody responses

An IFA assay was used to determine anti-PEDV IgG in the serum samples. As anticipated, the control group (non-vaccinated and non-infected) lacked a detectable humoral response to the virus. An FFN assay was used to evaluate neutralizing anti-PEDV immunoglobulin in milk samples obtained 48 hours postpartum. As expected, the control group lacked detectable anti-PEDV antibodies in their milk.

Unit A animals were non-infected and had been iPED+ vaccinated twice at a 3-week interval, with the last dose administered 3 weeks before farrowing (Figure 1A). As shown in Table 1, serum samples obtained 3 weeks post partum (approximately 6 weeks after the last vaccination) had anti-PEDV IgG reciprocal titers that ranged

from undetectable (< 20) in four sows to 80 in one of the six animals. Similarly, their 48-hour post-farrowing milk samples had neutralizing antibody titers ranging from undetectable in three to 80 in one sow.

Animals in two separate production units (B and C) were non-vaccinated prior to experiencing a PEDV outbreak. Only oral immunization by feeding back intestines from euthanized moribund neonates was performed in both units following the outbreaks (Figure 1B and 1C). In Unit B, one oral immunization was performed within days after the outbreak, which was approximately 4.5 months prior to farrowing. None of the six sows had detectable anti-PEDV IgG serum antibodies at the time of farrowing (Table 1). In contrast, their milk samples contained antiviral neutralizing antibodies, ranging in titers from 160 to 1280. Unit C sows had received three consecutive oral immunizations starting 24 hours after their outbreak, which was approximately 3.5 months prior to farrowing. In this case, all animals at the time of parturition had serum anti-PEDV IgG titers ranging from 20 to 160 and milk neutralizing-antibody titers ranging from 320 to 2560 (Table 1).

Unit D animals experienced one natural PEDV outbreak that initiated in the farrowing unit within 2.5 months prior to parturition of the third group of sows. This third group of sows had three iPED+ vaccinations approximately 12 and 9 weeks and 1 week prefarrowing, as well as two oral immunizations (intestinal contents of known Ct value) administered 8 and 5 weeks prior to farrowing (Figure 1D). A total of five serum samples were obtained from each study animal, starting 1 day prior to oral immunization, and weekly thereafter, with the last one obtained a few days prior to parturition. The highest serum anti-PEDV IgG titer was ≥ 320 for four sows on the first sampling, and was maintained in only one sow for the next two tests (Table 2). In general, humoral immunity inconsistently fluctuated and tended to decrease over the 5-week period, even following oral immunization. The milk antiviral neutralizing-antibody titers ranged from 80 to 1280 and did not necessarily correlate with the serum IgG titers in the last week serum samples were collected, a few days prior to parturition.

Table 1: Serum and milk anti-PEDV titers in sows in three independent swine facilities, units A, B, and C*

Sow	Group A		Group B		Group C	
	Serum†	Milk‡	Serum†	Milk‡	Serum†	Milk‡
1	80	Neg	Neg	320	160	2560
2	Neg	Neg	Neg	160	80	2560
3	Neg	80	Neg	1280	80	640
4	20	40	Neg	1280	20	320
5	Neg	Neg	Neg	320	40	320
6	Neg	20	Neg	320	80	1280

* Units B and C had experienced outbreaks of porcine epidemic diarrhea, while Unit A remained naive. Serum and milk samples were collected 48 hours post partum. Group name indicates the farm of origin.

† Serum anti-PEDV IgG titers were determined by an indirect fluorescent antibody (IFA) assay. Samples were tested in duplicate using twofold dilutions from 1:20 to 1:320. Titers are given as the reciprocal of the highest dilution of a sample in which a detectable anti-PEDV IgG result was obtained. Negative (Neg) result indicates that anti-PEDV IgG was not detected at the 1:20 dilution.

‡ Milk anti-PEDV neutralizing Ig titers were measured by a fluorescent focus neutralization (FFN) assay. Titers are given as the reciprocal of the highest dilution of a milk sample in which a detectable result was obtained.

PEDV = porcine epidemic diarrhea virus.

PEDV shedding

The relative amount of PEDV shed in the feces was temporally determined in Unit D, as units B and C were not available for such testing. Natural PEDV infections occurred in Unit D in the two previously pregnant groups of animals at their time of farrowing. Subsequently, the third group of pregnant sows was orally immunized 8 and 5 weeks prior to farrowing, as described. The second and third outbreaks were attributed to the oral immunization procedure as well as likely residual environmental contamination. Overall, virus shedding was not consistent over the 5-week prior (Table 2). Only low levels of virus were sporadically detected in seven of the nine sows during the five time points, with Ct values ranging from 36 (positive) to 39 (suspect). Three of the seven sows were in the suspect range, with one animal yielding three and the other two sows only one such result. The other four sows had one positive Ct value each, but were otherwise negative; for two of these sows, their first detectable fecal shedding was 2 to 3 days prior to parturition. When PEDV is being shed in the feces just prior to farrowing, the neonates are at risk of infection, regardless of the level of colostral protection they may have received. In fact, the neonates did succumb to PEDV infection (as determined by PEDV rRT-PCR), albeit with milder disease than the previous two litters.

Follow-up to case study

Unit A remained PEDV-free. Unit B did not experience further PEDV outbreaks in the batch farrowings subsequent to oral immunization. Similarly, after the oral immunization protocol, Unit C continued on a successful path of PEDV eradication 9 weeks post introduction. Unit D was depopulated due to the rRT-PCR-detectable environmental levels of PEDV that remained despite the rigorous decontamination attempts, and because of the change of plans for the unit (depopulation with repopulation).

Discussion

This case study was undertaken to provide data on whether parenteral iPED+ (first generation) vaccination alone was adequate to illicit detectable milk anti-PEDV antibodies, or if oral immunization with infectious PEDV was required. It is well-documented that porcine neonates rely upon passive immunity obtained from the colostrum and milk of their dam, as there is negligible placental transfer of antibodies during gestation.[7] Nearly all the colostral IgG and IgM and only 40% of the IgA originates in the systemic circulation of the dam; the change to mammary tissue origin occurs later in lactation.[8] During the first 48 hours of lactation after parturition, the Ig content of

Table 2: Serum and milk anti-PEDV reciprocal antibody titers and results of PEDV rRT-PCR on fecal samples in Unit D*

Sow	Serum† (fecal swabs‡)					Milk§
	0 weeks	1 week	2 weeks	3 weeks	4 weeks	
1	≥ 320 (39.8)	160 (39.0)	40 (38.10)	Neg (Neg)	Neg (Neg)	320
2	≥ 320 (Neg)	≥ 320 (Neg)	160 (36.9)	80 (Neg)	160 (Neg)	640
3	≥ 320 (Neg)	≥ 320 (38.7)	160 (Neg)	160 (Neg)	160 (Neg)	80
4	≥ 320 (Neg)	≥ 320 (Neg)	≥ 320 (Neg)	160 (Neg)	160 (Neg)	1280
5	80 (Neg)	40 (Neg)	40 (Neg)	20 (Neg)	80 (35.6)	640
6	80 (Neg)	20 (Neg)	40 (34.5)	Neg (Neg)	20 (Neg)	160
7	160 (Neg)	80 (Neg)	20 (Neg)	Neg (Neg)	Neg (Neg)	320
8	160 (Neg)	40 (38.4)	40 (Neg)	Neg (Neg)	40 (Neg)	1280
9	80 (Neg)	20 (Neg)	40 (Neg)	20 (Neg)	Neg (36.8)	320

* Serum and fecal swabs were obtained from sows prior to oral inoculation (0 weeks) and weekly thereafter for four additional time points, with the last samples collected a few days prior to parturition.

† Serum anti-PEDV IgG levels of sows were determined by an indirect fluorescent antibody assay. Samples were tested in duplicate using twofold dilutions from 1:20 to 1:320. Titers are given as the reciprocal of the highest dilution of a sample in which a detectable anti-PEDV IgG result was obtained. Negative (Neg) result indicates that anti-PEDV IgG antibodies were not detected at the 1:20 dilution.

‡ PEDV rRT-PCR Ct values for fecal swab samples are provided: positive (Ct value ≤ 37), suspect (Ct value, 37.01 to 40), or negative (Neg), Ct value > 40).

§ Neutralizing antibodies to PEDV in milk were measured using a fluorescent focus neutralizing assay. Samples were considered negative if antibodies were undetectable at a 1:40 dilution.

PEDV = porcine epidemic diarrhea virus; rRT-PCR = real-time reverse transcriptase polymerase chain reaction; Ig = immunoglobulin; Ct = cycle threshold.

colostrum or milk is very high, with IgG in the highest relative concentration followed by IgA then IgM (ratio of 76:17:7).[9]

Unit A sows received only iPED+ vaccinations at 6 and 3 weeks pre-farrowing. Both their serum IFA IgG and milk anti-PEDV neutralizing antibody levels were low, with titers of 80 or less. Unit B sows were orally immunized once approximately 4 months prior to their subsequent batch farrowings. Although the milk titers in these sows ranged from 160 to 1280, all serum titers were negative. The serum results may reflect an empirical observation that serum anti-PEDV antibodies do not appear to persist post infection. Similarly, in Unit C, sows were orally immunized on 3 successive days after their initial PEDV outbreak. Following this more rigorous immunization approach, the animals that farrowed nearly 3 months later had the highest milk anti-PEDV neutralizing antibody titers of the three units, with all six study sows having detectable serum IgG. These outcomes indicate that oral immunization induced higher levels of milk anti-PEDV-neutralizing antibodies than parenteral vaccination with PEDV antigen alone. However, the protective passive antibody titer for neonates has yet to be determined.

The Unit D sows in the study were provided with parenteral iPED+ vaccination at 9 and 2 weeks prior to farrowing, as well as oral immunization at 8 and 5 weeks pre-farrowing. The temporal serum sampling of the sows revealed that overall anti-PEDV antibody titers wane fairly rapidly (Table 2). As for TGEV, serum antibodies may not provide significant protection against PEDV infections.[3,4] Empirical information relates that feedback of neonatal intestines would stop a TGEV outbreak in a herd for the remainder of the "TGEV season." This has not been the case for PEDV. After feedback, a herd may be re-infected and manifest clinical disease months later in the same "season." Such empirical information may suggest the possibility that PEDV has immunosuppressive properties not evident in TGEV; such a hypothesis has yet to be examined. It is known that stimulation of GALT by oral inoculation or natural exposure to virulent virus achieves the most effective protective anti-TGEV immunity.[3] The active immunity that results from such enteric replication of the virus also involves induction of cell-mediated immunity (CMI), as well as production of intestinal secretory IgA.[10] The GALT CMI to TGEV was found to persist for only 14 days after oral inoculation in neonates (7 days of age) and lacked a natural killer cell component (part of innate immunity) in pigs of this age. In comparison, the GALT CMI persisted at least 110 days in pigs 6 months of age.[3] This reflects the noted age-dependent resistance to TGEV infections.[10,11] Such age resistance does not appear to be associated with PEDV infection, as the virus is known to cause disease in adults.

Although serum immunoglobulins are the initial source of colostrum and milk immunoglobulins, the measured milk anti-PEDV neutralizing-antibody titers did not necessarily correlate with the serum antiviral IgG titers, specifically the last samples collected a few days prior to parturition. This is also evident between units in this study in that while serum anti-PEDV IgG titers for Unit D were highest, their milk antiviral neutralizing antibodies were not. Such lack of correlation may be real or simply reflect a sensitivity disparity in the type of antibodies the two assays are detecting (IFA IgG detecting any anti-viral antibody; FFN detecting only neutralizing antibodies). From the perspective of the neonate, one would consider the milk anti-PEDV neutralizing antibody titers to be critical.

Furthermore, at the time of parturition, sows in the Unit D farrowing unit were still shedding PEDV in their feces (25 days after their last oral immunization). This is not unexpected, as neonates that recovered from a natural PEDV infection under field conditions shed the virus in their feces for up to 56 days post infection.[12] Interestingly, neonates of the Unit D sows did not manifest clinical disease until about 5 days of age and their disease was much milder than that during the initial PEDV outbreak in this herd. Therefore, passive immunity appeared to offer a level of protection, but eventually was overwhelmed. Although the duration of PEDV shedding from such infected adult animals may vary, it would be prudent to perform oral inoculations at least 40 days prior to parturition and in a different location than the farrowing unit.

Implications

- Under the conditions of this case study, oral immunization may be the more efficacious PEDV immunization modality
- There appears to be a lack of correlation between milk anti-PEDV neutralizing antibody titer and serum antiviral IgG titer.

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Conflict of interest

None reported.

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Reference values for immunocrit ratios to assess maternal antibody uptake in 1-day-old piglets

Brianna M. Peters, BS; Seth A. Krantz, DVM; Derald J. Holtkamp, DVM, MS; Chong Wang, PhD; Lucina Galina Pantoja, DVM, PhD

Summary

Colostrum intake is an essential component for piglet survival. Assessing colostrum intake, and consequently transfer of immunoglobulins (Igs), has been difficult to quantitate in swine. In this study, the authors first sought to subjectively determine the least stressful method to collect necessary sample quantities for Ig quantitation. The immunocrit ratio (IR) method was used to quantify a benchmark Ig level for a commercial production system. Lastly, the authors sought to identify associations between IR and production parameters. The cephalic vein provided consistent sample volumes and caused minimal animal distress. Additionally, a small volume of serum (30 µL) can be used for IR testing. An IR benchmark was determined to be 0.098 for this production system. For this study, no significant associations were found between pre-weaning mortality or average daily weight gain and IR. Birth weight and parity had significant effects ($P < .05$) on IR, with parity 1 litters having lower IR than higher parity litters. Using the IR technique to identify IR benchmarks for piglets will help producers improve colostrum intake opportunities in piglets with suboptimal Ig levels. The IR method ascertains whether piglets are receiving adequate maternal antibodies until their own immune systems are developed.

Keywords: swine, colostrum, on-farm, immunocrit, immunity

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Resumen - Valores de referencia para los índices de inmunocrito para evaluar el consumo de anticuerpos maternos en lechones de un día de edad

El consumo de calostro es un componente esencial para la supervivencia del lechón. La evaluación del consumo de calostro, y por ende la transferencia de inmunoglobulinas (Igs por sus siglas en inglés), ha sido difícil de cuantificar en cerdos. En este estudio, primero, los autores investigaron como determinar subjetivamente el método menos estresante para recolectar la cantidad de muestra necesaria para la cuantificación de la Ig. Se utilizó el método de índice de inmunocrito (IR por sus siglas en inglés) para cuantificar un nivel comparativo para la Ig para un sistema de producción comercial. Segundo, los autores buscaron identificar asociaciones entre el IR y los parámetros de producción. La vena cefálica proporcionó volúmenes de muestra consistentes y causó mínima molestia al animal. Además, un pequeño volumen de suero (30 µL) puede utilizarse para pruebas de IR. Se determinó un punto de referencia de IR de 0.098 para este sistema de producción. En este estudio, no se encontraron asociaciones significativas entre la mortalidad predestete o la ganancia media de peso diaria y el IR. El peso al nacimiento y la paridad tuvieron efectos significativos ($P < .05$) en el IR, las camadas de paridad 1 tuvieron un IR más bajo que las camadas de paridad más alta. La utilización de la técnica IR para identificar los puntos de referencia IR para lechones ayudará a los productores a mejorar las oportunidades de consumo de calostro en lechones con niveles de Ig subóptimas. El método IR comprueba si los lechones están recibiendo anticuerpos maternos adecuados hasta que su propio sistema inmunológico se desarrolle.

BMP: College of Veterinary Medicine, University of Tennessee, Knoxville, Tennessee.

SAK: Tosh Farms, Henry, Tennessee.

DJH, CW: Department of Veterinary Diagnostic and Production Animal Medicine, College of Veterinary Medicine, Iowa State University, Ames, Iowa.

LGP: Zoetis, Florham Park, New Jersey.

Corresponding author: Dr Lucina Galina Pantoja, Zoetis, 100 Campus Drive, Florham Park, NJ 07932; Tel: 615-545-4292; E-mail: lucina.galina@zoetis.com.

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Peters BM, Krantz SA, Holtkamp DJ, et al. Reference values for immunocrit ratios to assess maternal antibody uptake in 1-day-old piglets. *J Swine Health Prod.* 2016;24(1):36–41.

Résumé - Valeurs de référence pour les ratios d'immunocrites afin d'évaluer l'absorption d'anticorps maternels chez des porcelets âgés de 1 jour

La prise de colostrum est un élément essentiel pour la survie des porcelets. L'évaluation de la prise de colostrum, et par conséquent le transfert d'immunoglobulines (Igs), a été difficile à quantifier chez le porc. Dans la présente étude, les auteurs ont premièrement visé à déterminer subjectivement la méthode la moins stressante pour prélever les quantités nécessaires d'échantillon pour la quantification d'Ig. La méthode du ratio d'immunocrite (IR) a été utilisée afin de quantifier un niveau étalon d'Ig pour le système commercial de production. Finalement, les auteurs ont voulu identifier les associations entre IR et les paramètres de production. L'utilisation de la veine céphalique a permis d'obtenir des volumes constants d'échantillons et causait un minimum de détresse chez les animaux. De plus, un faible volume de sérum (30 µL) peut être utilisé pour l'épreuve d'IR. Une valeur étalon d'IR de 0,098 fut déterminée pour ce système de production. Pour la présente étude, aucune association significative ne fut établie entre la mortalité pré-sevrage ou le gain de poids journalier moyen et l'IR. Le poids à la naissance et la parité avaient des effets significatifs ($P < 0,05$) sur l'IR, les portées de parité 1 ayant un IR plus faible que les

portées de parités plus élevés. En utilisant la technique d'IR pour identifier les balises d'IR pour les porcelets aidera les producteurs à améliorer les opportunités de prises de colostrum chez les porcelets avec des niveaux suboptimaux d'Ig. La méthode d'IR détermine si les porcelets reçoivent des quantités adéquates d'anticorps maternels jusqu'à ce que leur propre système immunitaire soit développé.

Passive immunoglobulin (Ig) uptake via colostrum ingestion is of paramount importance for development of a newborn piglet's immune system. Colostrum contains antibodies and cells, such as macrophages and B and T lymphocytes, involved in both innate and humoral immunity.[1-3] Compared to older animals, a newborn piglet's immune system has a limited capacity to synthesize Igs for the first 3 to 6 weeks of life.[4] A piglet's ability to uptake these constituents influences its survival during the first several weeks of life, as these components are the newborn piglet's only source of protection against pathogens.[5,6] In studies evaluating the effect of time on the Ig content of sow colostrum, the highest concentration of IgG in colostrum is seen at the start of parturition. After 24 hours, the level decreases to basal levels for sow milk.[7] While the industry does not currently have a means of inducing continuously high globulin concentrations in sow milk, it is possible to implement colostrum management practices in order to promote optimal nursing during the time when piglet enterocytes allow peak globulin absorption. However, to date, it has been difficult to routinely quantify Igs to assess the need for colostrum management. Even though cattle and swine share the same mechanism of maternal immunity, until 1980, there was limited research regarding the quantification of swine Ig transfer.[8,9] To amend this oversight, Yaguchi et al[8] used technologies such as refractometry, electrophoresis, and spectrophotometry. In an attempt to make Ig quantitation more accessible to producers, a sandwich enzyme-linked immunosorbent assay (ELISA) was developed in 1985.[10] Unfortunately, this ELISA still requires 3 hours of incubation time post sample collection. Recently, Vallet et al[11] developed a simple, rapid, and inexpensive method to measure passive transfer of Igs from dam to piglet. Vallet et al[11] tested serum samples from 1-day-old piglets via the immunocrit method to generate an immunocrit ratio (IR). In the immunocrit procedure, aliquots of serum and 40% ammonium sulfate solution are mixed to precipitate the Ig proteins present. The length of precipitate formed in the hematocrit tube is then compared to the residual serum solution in the tube to calculate the IR as a decimal. Using this method, a research swine herd IR benchmark was established in early 2013.[11] However, benchmark immunocrit values and the ability to assess colostrum management with IR in commercial situations have not been validated. Therefore, the three objectives of this study were to evaluate alternative sampling techniques in neonatal piglets for the IR method, generate a benchmark for a desirable IR value in commercial populations, and determine whether an association exists between IR and production parameters such as wean weight and pre-weaning mortality.

Materials and methods

All animals in this study were raised and handled on commercial farms that are Pork Quality Assurance Plus certified and adhere to standards set forth by the National Pork Board. Due to the minimally invasive procedures utilized, an Institutional Animal Care and Use Committee protocol was not required.

The sows in the nine farms in this study were either C29 or 1070 PIC females (PIC, Hendersonville, Tennessee). These sows were housed in farrowing stalls, shared a common feed source, and were naive to porcine reproductive and respiratory syndrome virus. Pre-weaning mortality on these farms for the previous 6-month period ranged from 8% to 12%.

Sample collection

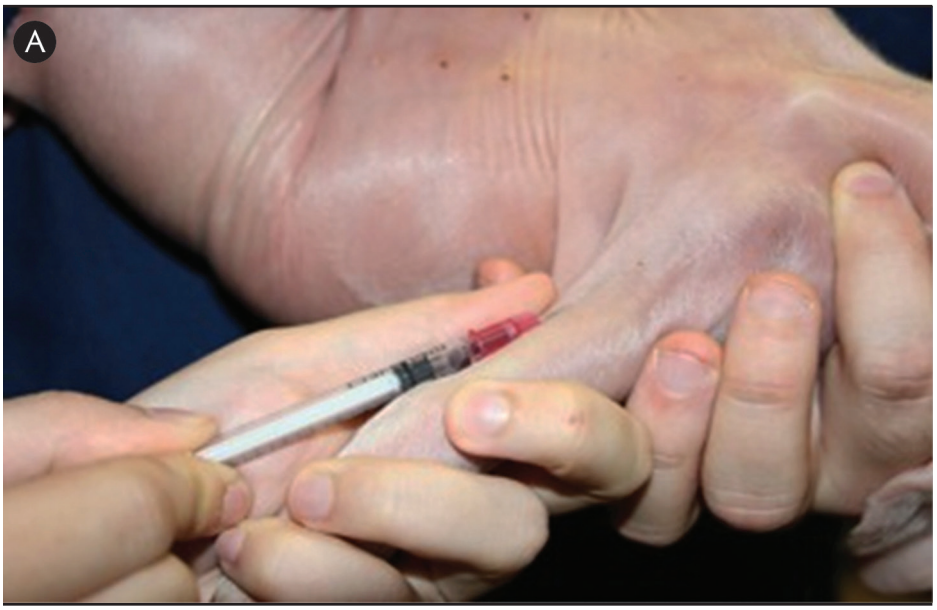
Blood samples for this study were obtained from piglets in all nine sow units within the same commercial production system. To first determine the most effective sampling method, a sample size of seventeen 24-hour-old piglets (± 12 hours) were selected from various litters within the same farrowing room. Selection of piglets was based on the ease of access to handlers. These piglets were assigned to various blood collection techniques via the cephalic vein or the medial or lateral saphenous veins. Blood collection via tail docking was attempted on several piglets from each of these groups. Blood collection at the time of tail docking would have provided the most practical collection method. Ideally, blood that pooled at the site of tail docking would have been suctioned into a 1-mL syringe. Unfortunately, an insufficient amount of sample (blood) pooled at the docking site. Using a 1-mL syringe and a 22-gauge × ¾-inch needle, 1 mL of blood was collected from each piglet to ensure an adequate volume (30 µL) of serum for testing. Piglet sample group size (n = 17) was determined by convenience while factoring in venous access and practicality for producers (no power studies were performed to determine this sample size). In order to not disrupt the lactation cycle of the litter, no more than two piglets were selected from the same sow. The degree of stress experienced by the animal was subjectively evaluated on the basis of the authors' observations of subject vocalization and attempts to avoid restraint. A technique was considered successful if 1 mL of blood was collected from at least three piglets from the sample group of 17.

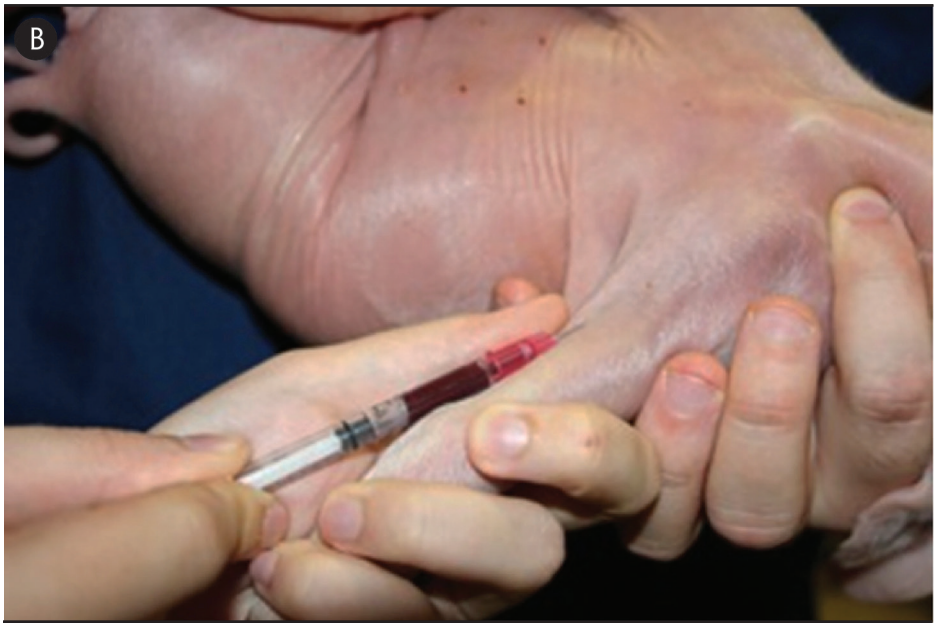
Once the successful sampling technique was found (via cephalic vein), 1 mL of blood from a new selection of 17 piglets was collected to determine serum volume necessary (30 or 50 µL) to evaluate IR results. Requiring less sample volume would prove beneficial in cases where a veterinarian wished to evaluate colostrum intake in a particularly dehydrated or unthrifty piglet.

On the basis of these results, blood collection via the cephalic vein was chosen for the larger study in which 779 piglets were sampled.

For the larger study, 30 litters were sampled from each of the nine sow farms. A light (< 1.25 kg), medium (1.25 to 1.75 kg), and heavy (> 1.75 kg) piglet was chosen, on the basis of birth weight, from each trial sow per farm. Sex, birth weight, sow parity, birth dam's ID, total born alive in the litter in which the piglet was born, time of birth (AM or PM), farrowing date, wean weight, and litter mortality were recorded for each piglet. Because previous research has stressed the importance of timing and piglet age with regard to passive Ig transfer, piglet birth time was approximated.[7] The birth-time designation was determined as follows: any piglet born between 7 AM and 2:59 PM was designated an AM piglet. This designation signified that caretakers were present to observe parturition and assist with drying and nursing if deemed necessary. A PM birth was assigned to any piglet born between 3 PM and the following morning

Figure 1: Illustration of a technique to draw blood from the cephalic vein using a 22-gauge, 3/4-inch needle to minimize stress to 1-day-old pigs. One mL of blood was more than sufficient to obtain a final volume of 50 µL serum, which can be used for the immunocrit ratio test to monitor colostrum intake.





at 7 AM when workers returned. Adjusted wean weights were calculated by multiplying average daily gain (kg) by 21 days. Blood (1 mL) was collected via the cephalic vein of piglets at approximately 24 hours of age by using a 22-gauge × ¾-inch needle and a 1-mL syringe (Figure 1). Samples were processed as previously described by Vallet et al.[11] Briefly, blood was allowed to clot overnight. Serum was separated and centrifuged at 1350*g* for 10 minutes. Next, 50-µL serum samples were mixed with 50 µL of 40% ammonium sulfate ($[NH4]_2SO_4$) in dolphin-nosed tubes. Using hematocrit microcapillary tubes, the mixture was centrifuged at 14,800*g* with the following adjustment in procedure: serum centrifugation time was increased from 5 to 10 minutes, as recommended to improve serum separation).[12] All blood samples were processed within 24 hours of collection.

Statistical analysis

A paired *t* test was performed on the IRs of 17 piglets to first determine the serum sample size (30 or 50 µL) needed to provide comparable results to the volume (50 µL) previously validated by Vallet et al.[11] For this study, the piglet was the experimental unit. Descriptive statistics, including mean, standard deviation (SD), and maximum and minimum values, were calculated for IR by farm, and scatter plots of IR were generated. Mean IRs were calculated for each farm by parity (P1, P2, P3, P4+), birth weight (< 1.25 kg, 1.25 to 1.75 kg, and > 1.75 kg), and adjusted wean weight (< 5 kg, 5 to 7 kg, and > 7 kg). For each farm, the average IR and SD were calculated.

The effect of IR on average daily gain (ADG) was analyzed using a mixed linear regression model with IR, birth weight, AM or PM birth, sex, parity, and total born alive as fixed effects and farm and sow as random effects. Birth weight was modeled as a continuous variable. The effects of IR, AM or PM birth, sex, parity, and total born alive on pre-weaning mortality were analyzed using a logistic regression model with the GLIMMIX procedure (SAS 9.2; SAS Institute, Cary, North Carolina). Values of $P < .05$ were considered significant. To determine a practical IR sample size, the SD for this production system (0.026) was used in addition to estimating calculated precision (distance from the mean to limit), based on a 95% confidence limit.

Results

Sampling technique

The jugular and vena cava veins were excluded as potential blood collection sites due to the difficulty in consistent accurate venipuncture of these vessels in neonatal pigs. Tail docking, a normal production procedure commonly performed at 5 days of age, likewise proved to be an insufficient means of collecting a significant volume of serum to analyze. Sample collection from the saphenous veins yielded the desired quantity of blood (1 mL), but results were not easily replicated while attempting to humanely restrain the animal. The cephalic vein provided consistent sample volumes and required minimal restraint and distress to the animal.

There was no significant difference in resulting IR values (mean ± SD; n = 17) when comparing 30-µL (0.113 ± 0.018) and 50-µL (0.114 ± 0.018) samples ($P = .37$). While a volume of 0.5 mL blood per piglet was determined to provide a sufficient amount of serum for the immunocrit test, a 1-mL sample was consistently collected for each piglet in this trial in order to account for potential retesting or extreme results.

Commercial production system benchmark

The target number of 90 blood samples collected per farm was reached for most sow farms (farms 1, 2, 3, 5, 6, and 9). However, in farms 4, 7, and 8, that target sample size was not achieved (actual sample size was 89, 69, and 81, respectively) because those sow farms had smaller sow-herd inventories and fewer farrowings occurring in a given week. Therefore, a total of 779 blood samples were obtained for immunocrit ratio measurements in this production system. A mean IR of 0.098 ± 0.026 was found for the entire operation (Table 1). Figure 2 provides an example of the uniformity a producer should strive to create within a herd. Farm 1 had the highest mean IR values and lowest variability (Figure 2A). Farm 9 consistently had the lowest IR values (Figure 2B). Farm 7 had mid-range IR values and the highest variability (Figure 2C). Across these three farms (1, 7, and 9) and the other farms (data not shown for farms 2 through 6 and Farm 8), it was demonstrated that small piglets can achieve a high IR and that heavier birth weight piglets may have lower IRs. In five of nine sow farms (56%), P1 litters had the lowest IRs when compared with older parity litters, reinforcing the importance of managing parity distribution in the herd. In six of nine farms (66%), the highest IR values were found in piglets with the heaviest birth weight and with heavier weaned weights. On the basis of the calculated precision, a sample size of 30 piglets (distance from the mean of 0.009) was found to be adequate to evaluate colostrum management strategies on farms, because 30 piglets generated the shortest distance from the mean to limit, compared with other sample sizes tested.

Table 1: Average immunocrit ratios (IR) in 779 blood samples from 24-hour-old piglets in nine commercial sow farms belonging to one swine production system*

Sow farm	Average IR (± SD)	Minimum	Maximum
1	0.116 (± 0.023)	0.047	0.158
2	0.101 (± 0.028)	0.016	0.148
3	0.099 (± 0.022)	0.046	0.172
4	0.098 (± 0.028)	0.015	0.156
5	**0.096** (± 0.022)	0.051	0.193
6	**0.095** (± 0.029)	0.056	0.129
7	**0.095** (± 0.029)	0.015	0.167
8	**0.093** (± 0.026)	0.015	0.138
9	**0.090** (± 0.025)	0.008	0.163
Production system	0.098 (± 0.026)	0.008	0.163

* Farms with IR averages lower than that of the whole production system (bold) were targeted for intervention to improve colostrum management. Immunocrit ratio measurement: 50 μL of serum was mixed with 50 μL of ammonium sulfate solution in a dolphin-nosed tube, then centrifuged in a hematocrit microcapillary tube for 10 minutes. The IR was calculated by dividing the length of the precipitate in the tube (mm) by the length of the entire sample (mm).

SD = standard deviation.

IR and production parameters

No significant associations were found between pre-weaning mortality or ADG and IR. Birth weight was the only significant independent variable for ADG ($P < .05$). Parity had a significant effect on IR ($P < .05$), with P1 litters having lower IR than higher parity litters.

Discussion

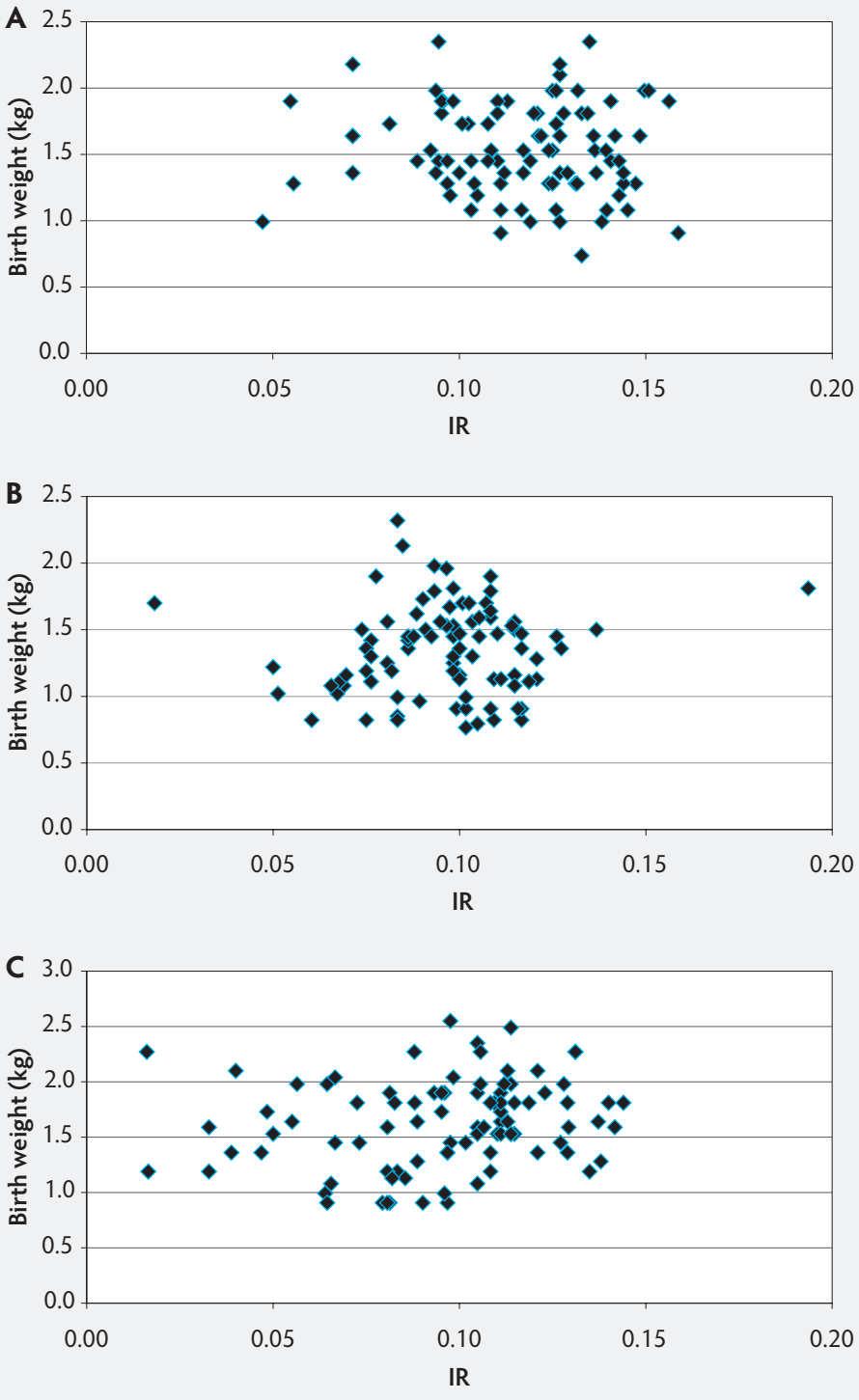
The first goal of this study was to determine a blood collection method that was less stressful to the piglet than methods used previously in research settings.[8, 11] The cephalic vein was easily accessed and repeatedly provided adequate sample volumes, and this method was minimally stressful to the animal. Consistent blood sample collection was expedited by adequate lighting, choosing well-hydrated piglets, and using experienced handlers. Hydration status was subjectively determined by assessing piglet body condition score and general attitude. If the needle slipped from the vein mid-draw, another method was used to collect the blood that pooled at the puncture site. Any contaminants, such as piglet moisture-absorbent powder, easily separated out upon centrifugation inside the hematocrit tubes.

For the immunocrit test, the paired *t* test results indicated that if a sample yielded less serum than anticipated, a smaller serum sample could be used. This validation will prove useful in situations when it is difficult to collect a sample from an animal, particularly in cases where piglets are severely dehydrated or in piglets with relatively low birth weights (< 0.68 kg).[13] Modification of the collection method and the volume of blood necessary increases handler awareness of piglet welfare.

Vallet et al[11] reported that an immunocrit ratio of 0.125 coincided with high piglet survivability in a research setting. This value was used as a desirable benchmark reference for the current study. Our data suggests that IR averages are lower in commercial settings. An explanation for this difference might be that the study of Vallet et al[11] had a larger sample size (number of animals) within a single farm. In the present study, there was a small sample size per farm. The average IR of 0.098 (± 0.026) for the entire production system was used as a benchmark in the current study. Colostrum management could be improved within this production system by focusing efforts on the four farms (farms 6, 7, 8, and 9) with the lowest IRs. The highest-ranking IR value (0.116 ± 0.023) was identified as an achievable benchmark for commercial operations.

Low birth weights did not appear to condemn pigs to poor colostrum intake (as evaluated by indirect measurement of immunoglobulins using IR), and improving and monitoring colostrum intake among all pigs, regardless of birth weight, may be beneficial. One possible method to provide equal suckling opportunity for pigs within a litter is split suckling. Split suckling is a technique used to minimize competition amongst littermates by allowing half of the litter to nurse colostrum for 1 to 2 hours and then allowing the second half of the litter to nurse. Additionally, caretakers should assess the likelihood of colostrum ingestion on abdomen fullness rather than relying solely on piglet birth weight. These findings suggest that the producer's goal, in addition to producing top-quality animals, should be to

Figure 2: Examples of immunocrit ratios (IR) in relation to piglet birth weight. Panel A: Farm 1 had consistently high IR values (most IR values are clumped together); Panel B: Farm 9 had consistently low IR values; and Panel C: Farm 7 had mid-range IR values and high variability.



achieve consistent Ig intake, with fewer than 15% of pigs below an IR of 0.116.

It was interesting to observe that all piglets born in the medium-range weight group (1.25 to 1.75 kg) had the lowest IR. This result could be due to the attention caregivers gave to the lighter and heavier piglets during split suckling and suggests that care also needs to be given to the medium-sized piglet.

Contrary to reports by Vallet et al,[11] a statistically significant association was not found between IR and pre-weaning mortality or ADG in this study of commercial animals. Despite this difference in findings, it is still a logical goal to strive to create a herd that is immunologically as uniform as possible. The antibodies that are garnered from colostrum ingestion still represent the piglet's sole source of defense against pathogens during the first 4 to 6 weeks of life. Ensuring adequate colostrum intake during the first 24 hours of life acts as a preventive measure against future infections and resulting production losses.

Parity was the only variable that impacted IR. Producers with high replacement rates might benefit from the knowledge that offspring of P1 sows appear to be at an immunological disadvantage. For producers with high replacement rates and concurrent average IRs below commercial values, P1 sow offspring should be the caretakers' primary focus in moving forward with colostrum-management improvement strategies.

The supply cost for performing an IR test is approximately $1.22 (US$) per piglet (2013 estimate). This accounts for the 1-mL syringe, a 22-gauge × ¾-inch needle, hematocrit tube, and blood tube needed per sample. Additional variable costs depend on the veterinarian's access to and quantity purchased of the following: a micropipette, micropipette tips, ammonium sulfate, a standard centrifuge, and a hematocrit centrifuge. Because a sample size of 30 piglets was adequate to evaluate colostrum management strategies on farms, a recommendation is to choose 1-day-old piglets from 10 different litters of mixed sow parities and select one light, one medium, and one heavy piglet per litter as a representative sample of the population. The immunocrit values in the commercial farms reported here can be used as benchmarks to monitor colostrum management practices.

Implications

- The cephalic vein is a reasonable site for collecting blood samples from neonatal pigs.
- A serum volume of 30 µL may be used for the described immunocrit method.
- A sample size of 30 piglets per farm, including one light, one medium, and one heavy piglet from 10 litters of different parities, is a good target size to survey colostrum management on a sow farm.
- Sampling piglets once or twice per year and comparing their immunocrit ratios to the herd average will help to assess

on-farm colostrum management. This system has the potential to attain an IR of 0.116. Individual production systems should ideally perform the immunocrit procedure in order to establish their system's goal IR.

- Under the conditions of this study, pigs from P1 litters have significantly lower IRs, which may place them at a greater immunological disadvantage.

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Conflict of interest

None reported.

Disclaimer

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National Pork Board announces Blue Ribbon Panel on antibiotics

The National Pork Board has announced members of its Blue Ribbon Panel on antibiotics, an outcome of the Pork Checkoff's stewardship plan first defined in June. The new third-party panel includes experts with specific experience in and knowledge of antibiotic practices or consumer marketing, but who are independent of National Pork Board practices.

"The critical role antibiotics play in pig farming is one of the most misunderstood facets of food production today," said Chris Hodges, National Pork Board chief executive officer. "We thank these leaders for their assistance and appreciate their range of expertise. From rigorous scientific study to food service and retail management, these experts will help us continue to build consumer trust and confidence in meat production."

For more information, go to **www.pork.org/antibiotics** or contact Jennifer Koeman at **JKoeman@pork.org** or 515-223-2633.

Checkoff starts new producer awareness campaign on new antibiotics rules, creates Antibiotics Resource Center

As part of the Pork Checkoff's year-long producer awareness and education campaign about the new rules affecting antibiotics in 2017, a comprehensive set of activities is now underway. According to Mike King, the Checkoff's director of science communications, these tactics include new fact sheets, brochures, newsletters, and an advertising campaign aimed at getting producers ready for the new Veterinary Feed Directive and the prescription rule for water-based medication.

For more information, see the Checkoff's Antibiotics Resource Center at **www.pork.org/antibiotics** or contact Mike King at **MKing@pork.org** or 515-223-3532.

New Employee Safety Toolkit available

Preliminary 2014 Bureau of Labor Statistic data show that the non-fatal injury and illness incidence rate for hog production is nine per 100 full-time workers. This rate is 2.8 times higher than all industry averages, 2.5 times higher than construction, and 1.6 times higher than crop production. To help improve these statistics, the Pork Checkoff is revising worker safety training materials to better address the hands-on, highly visual learning styles of barn workers. The new Employee Safety Toolkit will be available in early 2016 and will have a format similar to that of the Safe Animal Handling Toolkit.

The new kit will cover 21 key safety topics, ranging from hazardous gases to good housekeeping.

To accommodate different training environments and needs, the training materials will be flexible, interactive, and reflective of barn realities. The materials, available in both English and Spanish, will include video, PowerPoint presentations, supplemental knowledge checks, and practical skill-testing ideas.

For information about ordering the new toolkit, call the Pork Checkoff Service Center at 800-456-7675.

NPB news continued on page 45

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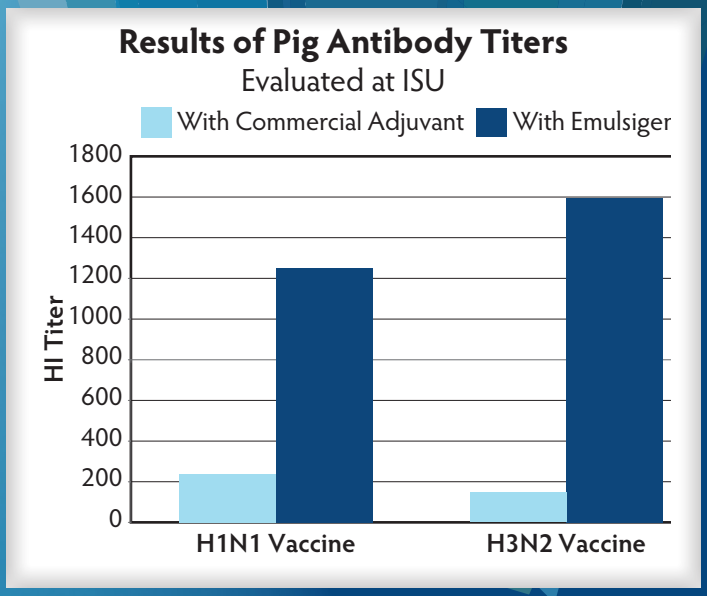

B.C. Lin, et al., AASV March 2006

NPB news continued from page 43

New infographic explains responsible antibiotic use on pig farms

The National Pork Board has debuted a new infographic depicting how US pig farmers work with their veterinarians to use antibiotics responsibly, helping to keep people, pigs, and the planet healthy.

"The role antibiotics play in pig farming is often misunderstood," said Chris Hodges, National Pork Board chief executive officer. "That's why we work closely with various groups in the food chain and why we're reaching out to consumers with information about how antibiotics are used on the farm. It's all part of our responsibility to build consumer trust in pork production."

To order free copies of the infographic, visit the Pork Store at **www.pork.org**.

Brent Sexton selected Alternate Student Delegate to AASV Board of Directors

The AASV Student Recruitment Committee is pleased to announce the selection of Brent Sexton (Iowa State University, 2018) as the incoming Alternate Student Delegate to the AASV Board of Directors.

Sexton is actively involved in a number of professional organizations and activities, including AASV and the Student Chapter of the AVMA. His veterinary-swine undergraduate work experience includes internships at Porcus Swine Veterinary in Odense, Denmark, and Harrisvaccines in Ames, Iowa. During the Porcus internship, he assisted with routine health consultations with swine producers, developed and implemented research techniques in clinical trials, compiled and analyzed data from those trials, and gained an understanding of European Union regulations and their effects on global agriculture. At Harrisvaccines, he learned the protocols associated with research and vaccine development, assisted with vaccine research and production, and formulated media for vaccine production.

This past summer, he participated in the Iowa State University Swine Veterinary Internship Program, conducting sow-farm biosecurity assessments with The Maschhoffs and Zoetis. He will be presenting the results of that research at the 2016 AASV Annual Meeting.

During 10 years of 4-H and FFA membership, he raised market, breeding, and show pigs and has served as the Iowa State Fair 4-H Swine Show Assistant Superintendent for the past 6 years. He has also attended and worked at the World Pork Expo for several years.

Following graduation, Sexton's career objective is to work in a multi-veterinary practice in rural Iowa. He would like to eventually buy into a veterinary business and become a managing partner.

Sexton assumes his duties as Alternate Student Delegate during the 2016 AASV Annual Meeting in New Orleans. The current alternate delegate, Emily Mahan-Riggs, will assume the delegate position currently held by Chris Sievers, who will rotate off the board. Emily and Brent will represent student interests within AASV as non-voting members of the board of directors and the Student Recruitment Committee.

Please join us in welcoming Brent to the AASV Board of Directors and thanking Chris for his service!

AASV debuts new display at National FFA Convention

For the eighth consecutive year, AASV members and staff promoted the swine veterinary profession at the National FFA Convention in Louisville, Kentucky. The AASV exhibit presented a fresh new look to convention-goers with the debut of a new, four-banner display. The eye-catching display features images of AASV members working with pigs and was designed for the association by graphic designer Tina Smith. The new display is easier to transport and quicker to set up than the previous AASV booth, which had been in service for more than 20 years.

Dr Todd Wolff coordinated the staffing of the display for the AASV Student Recruitment Committee, which oversees the association's participation in the convention. He was assisted by fellow AASV members Drs Angela Baysinger and Natalie Baker, as well as AASV's Director of Communication Dr Harry Snelson.

Over the course of 3 days, the AASV representatives visited with hundreds of high school and college students and their instructors about what it's like to be a swine veterinarian. They shared videos, posters, and information about swine diseases, biosecurity, and production practices, and passed out copies of AASV's recently revised "swine career brochure" to students interested in pursuing a career in veterinary medicine. "Ag" educators snapped up AASV's "advisor packet" of educational resources, exhausting the supply of 300 within the first 2 days of the convention.

The association expresses its appreciation to Joel Burkgren, who transported the display from the AASV office to Louisville and back, and assisted with setup and other logistical details associated with the exhibit.

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AASV FOUNDATION NEWS

Merck Animal Health provides funding for five $5000 scholarships

The AASV Foundation is pleased to announce that Merck Animal Health has provided $25,000 for a new veterinary student scholarship program. The AASVF-Merck Veterinary Student Scholarship Program seeks to identify and assist future swine veterinarians with their educational expenses.

Second- and third-year veterinary students enrolled in AVMA-accredited or -recognized colleges of veterinary medicine in the Canada, the Caribbean Islands, Mexico, South America, and the United States are eligible to apply for one of the five $5000 scholarships to be awarded. All applicants must be current student members of AASV. To apply, students must submit a resume and the name of a faculty member or AASV member to serve as a reference, along with written answers to four essay questions. Applications for the scholarships to be awarded in 2016 were due December 31, 2015.

The selection process will be conducted by a committee of four, which includes two AASV Foundation Board members and two AASV members-at-large. On the basis of the submitted materials, the student applicants will be scored and ranked on their past and current activities, level of interest in swine veterinary medicine, future career plans, and financial need. The five scholarship recipients will be announced during the 2016 AASV Annual Meeting in New Orleans, and the scholarship funds will be disbursed in March, after the conference.

The AASVF-Merck Veterinary Student Scholarship Program provides yet another opportunity for the AASV Foundation to fulfill its mission of "supporting the development and scholarship of students and veterinarians interested in the swine industry." For more information on scholarships and other AASV Foundation programs, see **www.aasv.org/foundation**.

MVP Laboratories donates raffle grand prize

For the past few years, the AASV Foundation raffle has proven to be a very successful fundraiser, and MVP Laboratories is doing its part to ensure its continued success. Last year, the company donated a 2015 Harley Davidson motorcycle for the raffle, which raised over $30,000 for the AASV Foundation!

This year, MVP Laboratories is once again donating the raffle grand prize, valued at $25,000. What is it? Take a look at **http://ecom.aasv.org/raffle** and see for yourself! While you're there, scroll down the page and purchase your chance to win this incredible prize! Tickets are $100 each, and can also be purchased from one of the auction committee members, or at the AASV registration desk during the AASV Annual Meeting in New Orleans.

The winning raffle ticket will be drawn and announced during the AASV Foundation Auction on Monday night, February 29, in New Orleans. The winner does not need to be present – so ALL AASV members can participate, knowing they have a chance to win and support the AASV Foundation at the same time. Thanks to MVP Laboratories, the AASV Foundation is a winner with every ticket purchased!

Jazz it up!

Give generously in the Big Easy!

Check out the items up for bid at **www.aasv.org/foundation** and make plans to participate in the 2016 AASV Foundation Live and Silent Auctions! Remember, if you're not attending the AASV Annual Meeting, you can submit bids by phone (515-465-5255) or e-mail (**aasv@aasv.org**) prior to February 23.

Since all of the items have been donated, the full amount of each winning bid will support AASV Foundation programs, including swine research, scholarships, swine externship grants, annual meeting travel stipends for students, tuition grants at the Swine Medicine Education Center, and more!

The auctions will be held Monday, February 29, during the AASV Annual Meeting in New Orleans. Thank you for supporting the AASV Foundation with your auction donations and bids!

AASV Foundation news continued on page 51

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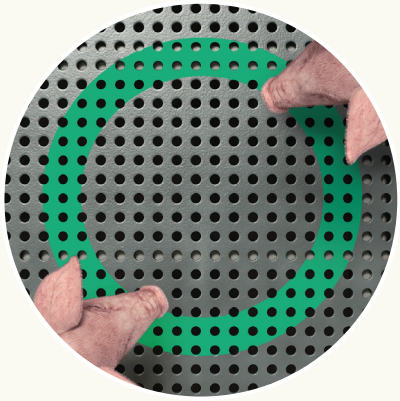

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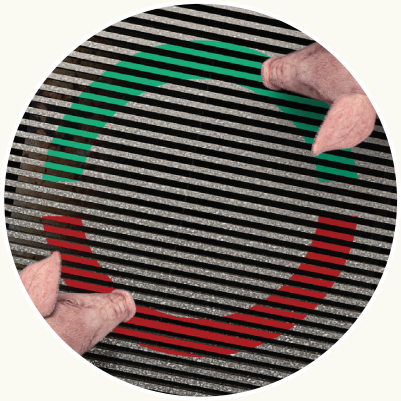

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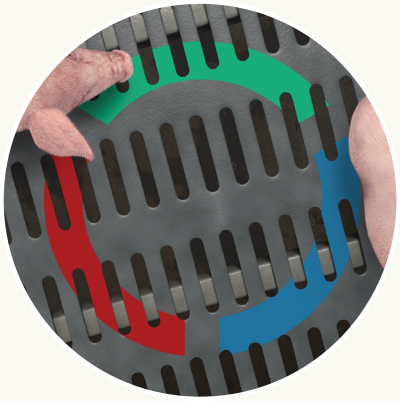

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AASV Foundation news continued from page 49

AASV Foundation issues call for research proposals: $60,000 available

As part of its mission to fund research with direct application to the profession, the American Association of Swine Veterinarians Foundation seeks research proposals for funding in 2016. Proposals are **due January 29, 2016,** and may request a maximum of $30,000 (US$) per project. A maximum of $60,000 will be awarded across two or more projects. The announcement of projects selected for funding will take place at the AASV Foundation Luncheon in New Orleans, Louisiana, on Sunday, February 28, 2016 (awardees may be notified in advance).

Proposed research should fit one of the five action areas stated in the AASV Foundation mission statement (see sidebar).

The instructions for submitting proposals are available on the AASV Foundation Web site at **https://www.aasv.org/foundation/2016/research.php**.

Proposals may be submitted by mail or e-mail (preferred).

A panel of AASV members will evaluate and select proposals for funding, on the basis of the following scoring system:

- Potential benefit to swine veterinarians/swine industry (40 points)
- Probability of success within timeline (35 points)
- Scientific/investigative quality (15 points)
- Budget justification (5 points)
- Originality (5 points)

For more information, or to submit a proposal:

AASV Foundation, 830 26th Street, Perry, IA 50220-2328; Tel: 515-465-5255; Fax: 515-465-3832; e-mail: **aasv@aasv.org**.

AASV Foundation Mission Statement

The mission of the American Association of Swine Veterinarians Foundation is to empower swine veterinarians to achieve a higher level of personal and professional effectiveness by

- Enhancing the image of the swine veterinary profession,
- Supporting the development and scholarship of students and veterinarians interested in the swine industry,
- Addressing long-range issues of the profession,
- Supporting faculty and promoting excellence in the teaching of swine health and production, and
- Funding research with direct application to the profession.

Swine veterinarians invited to apply for Hogg Scholarship

The American Association of Swine Veterinarians Foundation is pleased to offer the Hogg Scholarship, established to honor the memory of longtime AASV member and swine industry leader Dr Alex Hogg. Applications for the $10,000 scholarship will be accepted until February 1, 2016, and the scholarship recipient will be announced on Sunday, February 28, during the Foundation Luncheon at the 2016 AASV Annual Meeting in New Orleans.

The intent of the scholarship is to assist a swine veterinarian in his or her efforts to return to school for graduate education (resulting in a master's degree or higher) in an academic field of study related to swine health and production.

Dr Alex Hogg's career serves as the ideal model for successful applicants. After 20 years in mixed-animal practice, Dr Hogg pursued a master's degree in veterinary pathology. He subsequently became Nebraska's swine extension veterinarian and a professor at the University of Nebraska. Upon "retirement," Dr Hogg capped off his career with his work for MVP Laboratories. Always an enthusiastic learner, at age 75 he graduated from the Executive Veterinary Program offered at the University of Illinois.

The scholarship application requirements are outlined below and on the AASV Web site at **http://www.aasv.org/foundation/hoggscholarship.htm**.

Hogg Scholarship application requirements

An applicant for the Hogg Scholarship shall have

1. Five or more years of experience as a swine veterinarian, either in a private practice or in an integrated production setting; and
2. Five or more years of continuous membership in the AASV.

Applicants are required to submit the following for consideration as a Hogg Scholar:

1. Current curriculum vitae,
2. Letter of intent detailing his or her plans for graduate education and future plans for participation and employment within the swine industry, and
3. Two letters of reference from AASV members attesting to the applicant's qualifications to be a Hogg Scholar.

Applications and requests for information may be addressed to AASV Foundation, 830 26th Street, Perry, IA 50220-2328; Tel: 515-465-5255; E-mail: **aasv@aasv.org**.

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MERCK Animal Health

AASV Swine Health Committee considers Seneca Valley virus response

The American Association of Swine Veterinarians (AASV), National Pork Board (NPB), National Pork Producers Council (NPPC), and Swine Health Information Center (SHIC) have been working collaboratively with federal and state animal-health officials, food-safety inspectors, packing-plant officials, and laboratory diagnosticians to address the recent increase in Seneca Valley virus (SVV) cases. Historically, vesicles in pigs automatically led to a suspicion of foot-and-mouth disease (FMD). This suspicion is less clear-cut with the recent increase in positive SVV cases. Seneca Valley virus is clinically indistinguishable from the vesicular foreign animal diseases (FADs) of swine, including FMD, vesicular stomatitis (VS), and swine vesicular disease (SVD). Therefore, it is imperative that veterinarians and producers continue to respond to vesicular lesions as if they represent an FAD until proven otherwise. Remember, accredited veterinarians are required to report the presence of vesicular lesions in swine to animal-health officials.

The challenge, from a regulatory standpoint, is how to ensure prompt investigation of vesicular cases while not unnecessarily delaying movements of animals determined to be negative for an FAD. This involves state animal-health officials, United States Department of Agriculture's (USDA's) Animal and Plant Health Inspection Service, and Food Safety and Inspection Service (FSIS) personnel, as well as accredited veterinarians and packing-plant officials. As I am writing this article in November, USDA is drafting a guidance document to describe how they plan to address these challenges.

"Remember, accredited veterinarians are required to report the presence of vesicular lesions in swine to animal-health officials."

While we await guidance from USDA, the AASV Swine Health Committee (SHC), at the request of SHIC, evaluated the status of, and possible responses to, the recent SVV cases. The committee met by conference call on September 8, 2015, and provided up-to-date information regarding the most recent cases as well as the results of a PCR survey of oral-fluid samples conducted at both the Iowa State University Veterinary Diagnostic Laboratory and the University of Minnesota Veterinary Diagnostic Laboratory. Each laboratory retrospectively tested approximately 1000 oral-fluid samples from swine not reported to be exhibiting clinical signs indicative of SVV (acute lameness accompanied by vesicular lesions on the snout or coronary band or hoof or both) submitted to the diagnostic laboratory during the week of August 24, 2015. Samples submitted from numerous states tested PCR-positive.

The committee concluded that early evidence suggests SVV is a widespread emerging swine-production disease fitting the criteria of a TYPE 3 emerging disease outbreak. Those criteria include the following:

- Widespread areas of infection, and/or infections that are geographically and epidemiologically distinct, involving a large proportion of swine-production centers in the United States.
- There is inadequate knowledge about the disease, how it spreads, effective prevention and/or control measures, and risk pathways for disease entry and spread.
- There is little to no likelihood of controlling the disease using quarantine, stop movement, or depopulation, and no known or effective vaccine, treatment, or control strategies.
- It is expected to take greater than 1 year to develop the needed tools and information to mitigate negative effects of the disease on swine health and welfare, and producer profitability.

Note: the TYPE 1, 2, and 3 designations are derived from a draft Emerging Disease Response Plan under development through a joint effort involving AASV, NPB, NPPC, SHIC, and USDA.

Complacency in continuing to monitor for FADs could be devastating to the livestock industry of the United States. **The AASV SHC determined that observation of vesicles in pigs should continue to be treated as evidence of a potential FAD, necessitating the following activities.**

Herd veterinarian roles and responsibilities:

- Intensive surveillance for gross lesions and clinical signs (observe the pigs).
- Upon encountering a suspect case, the veterinarian should
  - Call the state or federal animal-disease control officials,
  - Stay at the site to await instructions from state or federal animal-health officials, and
  - Stop all human, vehicular, and animal movements.

Advocacy continued on page 55

INDICATIONS
For treatment/control of swine bacterial respiratory disease (swine bacterial pneumonia) associated with *Actinobacillus pleuropneumoniae, Pasteurella multocida, Salmonella Choleraesuis* and *Streptococcus suis*.

IMPORTANT SAFETY INFORMATION
People with known hypersensitivity to penicillin or cephalosporins should avoid exposure to EXCENEL RTU EZ. Do not use in swine found to be hypersensitive. Withdraw 4 days prior to slaughter.

EXCENEL® RTU EZ
(*ceftiofur hydrochloride*)
Sterile Suspension

RESIDUE FREE GUARANTEE

See Brief Summary of Prescribing Information on the next page.

zoetis™

Advocacy continued from page 53

- Once the disease has been determined to not be an FAD,
  - As with any clinically sick animal, SVV-positive animals exhibiting clinically-active lesions cannot be shipped to slaughter.
  - Once lesions are no longer active, the state animal-health official(s) should notify the slaughter plant and the USDA FSIS of the diagnostic findings before the animals are shipped to slaughter, if healing erosions are still present. The FSIS is currently working to determine what additional documentation may be necessary to verify FAD-negative status.

Producer roles and responsibilities:

- Do not move animals that are ill or exhibiting clinical signs, including clinically active lesions,
- Segregate or isolate affected animals on the site (if possible),
- Disclose and report movements leading up to and immediately surrounding the onset of clinical signs,
- Allow for sample collection and submission, and
- Communicate with state and local officials.

Additional guidelines are available in the *Procedures to Report a Suspected Foreign Animal Disease* document on the AASV website (`https://www.aasv.org/documents/FAD-Reporting.pdf`).

Harry Snelson, DVM
Director of Communications

Vice-presidential candidate

Scanlon Daniels

I am honored and deeply humbled to have been nominated to run for the office of vice president of the American Association of Swine Veterinarians (AASV). The association has always benefitted from the experience and knowledge of our past officers, and it would be a privilege to follow in their path. I request your vote in this election.

Not all issues important to our membership and association can be addressed in this space, so I encourage you to contact me if you have specific concerns or opportunities you feel the association needs to address. Many of our concerns are ongoing issues, like antibiotic use and how we care for and house swine. There will be different concerns in the future, such as new or re-emerging diseases.

In our geographic region, we have a lot of center pivot irrigation. This results in fields that are large circles instead of traditional squares or rectangles. Local and regional businesses have tapped into the multiple meanings of the phrase "full circle" and incorporate the term into their business name or marketing materials. It has caused me to reflect on some of the challenges our profession faces, and how some of them are the same challenges we have always faced, and some are new. For example, we seem to have come full circle with the porcine epidemic diarrhea virus (PEDV) challenge, and now it occupies the position of concern that transmissible gastroenteritis virus previously held. In addition, attributes of food production have always been of concern to society. Think Upton Sinclair's *The Jungle*,[1] the activities of animal rights activist groups, and the regulatory and legislative actions around antimicrobial use. These issues seem to ebb and flow or go in circles of low and high intensity.

Technology is a major influence on the circles that influence us. For example, the way we listen to music has undergone dramatic change in the last 35 years. When I was young, you had to listen to the radio for hours to hear your favorite song, or you had to buy the cassette tape for your Sony Walkman. Now we have satellite radio and the likes of Pandora where you can customize stations to your specific taste. The interest in different music genres goes through circles of popularity, while the way we listen has evolved in dramatic ways.

The way we communicate is another area that has seen many changes. Not long ago, I showed my kids a picture of a rotary phone. They could tell it was a phone, and asked a lot of questions about how it worked. In contrast to the past, we no longer have a landline at our house and rely solely on our cell phones and Internet connection for personal communication. About the only time we send a letter is for thank-you notes. In our business, bills are paid on line and other communication is by cell phone, text, or e-mail. We still have a fax line at our office, but it forwards documents to our e-mail instead of printing a hard copy. We still communicate using verbal and written means, but the way we do it has changed in dramatic ways.

In contrast, other technologies have been relatively static. The work boots I wear are essentially the same as those that my parents and grandparents used. Zippers, patented in 1893, work so well that I am betting they will still be in use for another 123 years. This contrast of change and status quo is driven by functionality. In other words, what works the best eventually dominates.

So how is technology going to influence the challenges our profession and association face?

1. Technology affects how we fight disease. It may be in the processes we use to raise pigs, or it could be a specific product like a vaccine. For example, batch farrowing is making a resurgence in our area as a response to the PEDV challenge and as a way to source larger numbers of "single-source" pigs. Vaccine technology can play a major part in the elimination of disease as it did for pseudorabies virus, or in control as with porcine circovirus type 2.
2. Technology influences how we communicate. Rather than guessing what people are interested in, tools like Google Analytics report in real time the information for which people are searching. Savvy influencers use this information to be more effective in communicating their message.
3. Technology influences the speed that we need to make change. Think "treadmill theory," where you have to adopt new technologies faster and faster over time to maintain your same competitive position.
4. Technology influences our "brand." It can enhance our efforts to promote our profession and association, or it can be used by detractors to take away our influence.

In very simple terms, for some challenges or opportunities, our choices are to hang onto our rotary phones or accept the new communication technology of the digital age. For others, we need to discern if the challenges are just circles of interest and can be handled using the technologies and practices we have used for years, like zippers and boots.

I ask for your vote and look forward to serving on the executive board of the AASV. We have a fabulous organization through the efforts of our members, staff, committee chairs, and board leadership.

Reference

1. Sinclair U. *The Jungle*. New York, New York: Knopf, Doubleday & Company; 1906.

James Kober

It is an honor and a privilege to be nominated to serve as vice president of the American Association of Swine Veterinarians (AASV). The AASV has been an integral part of my life since before graduation from veterinary school. I have attended 29 of the last 30 annual meetings. It is a wonderful organization and, frankly, the envy of many other veterinary groups. I have never experienced the camaraderie and willingness to share knowledge at any other veterinary meeting I have attended.

I grew up on a small grain, fruit, and livestock farm in West Michigan. Over time, we had less fruit and concentrated more on livestock. We started with a few sows and about 60 head of beef cattle. After a few years, my father decided to focus on hog production and divested of the fruit and cattle. We expanded the herd to 90 sows, farrowing four per week. The farm was more labor intensive than today's farms, but we had some distinct advantages. We were isolated in the fruit belt. The closest pigs were nearly 3 miles away. We utilized two-site production; our finisher barn was built on our second farm over 6 miles away. My dad was adamant that we drive to an isolated corner of the farmstead to change boots, wash the truck cab, then clean and wash the livestock trailer after EVERY load of hogs that was sold. I'm not sure we knew those habits constituted good biosecurity at that time. The lessons of hard work and animal care that my father instilled in me have been a solid and valuable foundation on which to build my career in swine practice.

After high school, I attended Michigan State University (MSU), where I earned both my bachelor and DVM degrees. Education in swine medicine was not prevalent at MSU, but I was blessed to have a wonderful mentor in Dr Brad Thacker. I spent nearly as much time visiting farms with Dr Thacker as I did in class. He taught me the essentials of swine medicine as well as the art of veterinary practice.

I completed the Executive Veterinary Program (EVP) at the University of Illinois in 1995. At that time, Iowa State gave graduate credits to EVP graduates. A small group of us continued on to a master of science program and I earned an MS degree in swine health and production in 1998. I then became board certified in Swine Health Management through the American Board of Veterinary Practitioners (ABVP).

My supportive and patient wife, Donna, is also a veterinarian. She earned her MBA degree and currently teaches practice management at two universities, as well as business consulting with private practitioners. We have three wonderful children: Ben, who is in his first year at the Kentucky College of Osteopathic Medicine; Sarah, who is in her final semester at Purdue University; and Amy, who is a first-year student at Kent State University.

My first job after veterinary school was at a mixed animal practice in southern Michigan. Swine made up 50% of that practice and nearly all of those farms utilized outdoor production. I then moved to central Indiana and practiced with Dr Max Rodibaugh, a great mentor and a leader in the industry. In 1993 I started my own practice in Holland, Michigan. I worked primarily with small family farms, many of which grew rapidly as the industry consolidated in the late 1990's and early 2000's. I call on fewer farms now, but see many more pigs.

I believe giving back through community service and serving organized veterinary medicine is important. I served as trustee in our church for over 10 years and was chairman of the building and grounds subcommittee. I was the swine representative on the board of directors of the American Board of Veterinary Practitioners (AVBP) for 7 years and am currently the chairman of the ABVP Foundation. Animal welfare is also very important to me. I have been on the Michigan Veterinary Medical Association animal welfare committee for several years. I enjoy presenting Operation Main Street lectures to various audiences across West Michigan. I also participated in the Swine Advocacy Program in Washington, DC, through the National Pork Producers Association.

The AASV is one of the most respected veterinary groups, making it truly enjoyable and satisfying to serve on AASV boards and committees. I am currently AASV's

representative to the Clinical Practitioners Advisory Council for the AVMA. I have been on the JSHAP editorial board as the practice-tip reviewer, then as a regular reviewer for the last 5 years. I have also served on the Pig Welfare Committee for several years and am now the chairman of that committee.

Our profession and industry will always need to be promoted, and we need to manage obstacles as they arise. Although the welfare front has been quiet for a few months, it is important that we continue to educate our professional colleagues and the public, as we are the best advocates for swine welfare. The assault on antibiotic use will continue and our expertise will be needed to educate both producers and consumers. Verifying that pork is a safe and wholesome protein source will take the efforts of all swine veterinarians and pork producers working together. There will continue to be disease issues, both old and new, where we will need our medical experience and expertise. The AASV will rise up to the challenge. The leadership and staff is second to none. I am humbled to be nominated for the office of vice president of AASV. I look forward to the challenges.

Guidelines for authors submitting manuscripts

Prepare the manuscript in Word using Times New Roman 12-point font, double-spaced throughout. Submit manuscripts to the Publications Manager.

Please include:

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- For all authors, names (first, middle initial, last), affiliations, and academic degrees beyond bachelor's level; and
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We will have your summary professionally translated into French and Spanish.

Editorial office

Karen Richardson, Publications Manager, *Journal of Swine Health and Production*; Tel: 519-856-2089; Fax: 519-763-3117; E-mail: **pub_mgr@aasv.org**.

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Figures and tables

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- Do not paste figures into the word-processing document containing the text of the manuscript. Submit them separately, eg, submit figures created in Excel as Excel files, and submit figures created in other programs as .eps files (ie, save as .eps files from within the program that created the figures).
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- Provide us with numerical data for all figures, including SD or SE for means.
- Supply brief but complete titles for tables and legends for figures. Explain in footnotes abbreviations used in tables, using symbols to identify footnotes.
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Measurements

The *Journal of Swine Health and Production* adheres, with a few exceptions, to the style of the American Medical Association.[1] A conversion chart is included at the end of the author guidelines document on the Web site at **http://www.aasv.org/shap/guidelines.pdf**. Please see the Web version of author guidelines for full details on journal requirements for submitted manuscripts.

Reference

1. Iverson C, Christiansen S, Flanagin A, JAMA and Archives Journals Staff, eds. *AMA Manual of Style: A Guide for Authors and Editors*. 10th ed. New York, New York: Oxford University Press. 2007.

Upcoming meetings

Banff Pork Seminar
January 12-14, 2016 (Tue-Thu)
Banff Centre
Banff, Alberta, Canada

For more information:
Ashley Steeple
Banff Pork Seminar
4-10 Agriculture-Forestry Centre
University of Alberta
Edmonton, AB T6G 2P5, Canada
Tel: 780-492-3651; Fax: 780-492-5771
E-mail: pork@ualberta.ca
Web: http://banffpork.ca

2016 Pig-Group Ski Seminar
February 3-5, 2016 (Wed-Fri)
Copper Mountain, Colorado

For more information:
Lori Yeske
Pig Group
39109 375th Ave, St Peter, MN 56082
Tel: 507-381-1647
E-mail: pyeske@swinevetcenter.com
Web: http://www.pigski.net

American Association of Swine Veterinarians 47th 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

World Pork Expo
June 8-10, 2016 (Wed-Fri)
Iowa State Fairgrounds, Des Moines, Iowa
Hosted by the National Pork Producers Council

For more information:
Alicia Newman
National Pork Producers Council
10676 Justin Drive, Urbandale, IA 50322
Tel: 515-278-8012; Fax: 515-278-8014
E-mail: newmana@nppc.org
Web: http://worldpork.org

Association for Applied Animal Andrology
June 24-26, 2016 (Fri-Sun)
Vinci Centre Interantional de Congres de Tours
Tours, France

For additional information:
Dr Steve Lorton
Tel: 608-206-1078
E-mail: info@animalandrology.org
Web: http://www.animalandrology.org

For additional information on upcoming meetings: https://www.aasv.org/meetings/

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Shy pig

Photo courtesy of Dr John Waddell

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