

THE 11TH MIKE WILSON SWINE RESEARCH DAY

JUNE 4, 2014

9:30 AM – 3:15 PM

Arboretum Center, University of Guelph



The 11th Annual Mike Wilson Swine Research Day

June 4, 2014

9:30 AM – 3:15 PM

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- 9:30 AM Registration and coffee**
- 10:00 AM Bob Friendship: Welcome**
- 10:05 AM Zvonimir Poljak: Epidemiology and swine disease control**
- 10:45 AM Andreia Arruda: Spatial and temporal PRRS patterns in Ontario**
- 11:00 AM Ming Fan: Fibre degradation enzyme development**
- 11:15 AM Paul Luimes: Sow feeding programs**
- 11:30 AM Jim Squires: Boar taint and genetic markers**
- 11:45 AM Keith Warriner: Salmonella control in the pork chain**
- Noon Lunch and posters**
- 13:30 PM Davor Ojkic: PED update**
- 13:45 PM Mackenzie Slifierz: Zinc and antimicrobial resistance**
- 14:00 PM Emily Miller: Nitrogen retention for gestating sows**
- 14:15 PM Melissa Wiseman: Liquid feeding DDGS to piglets**
- 14:30 PM Kathy Zurbrigg: Heart lesions and transport death**
- 14:45 PM Ira Mandell: Gender effects on carcass and meat quality**
- 15:00 PM Kees de Lange: Announcement of winning posters and closing remarks**

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Spread and control of influenza in swine populations: insight from analysis of infectious disease data

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Abstract

Infectious diseases are rapidly emerging in various animal populations. The mechanism of emergence is similar to the mechanisms in people. This typically includes crossing the species barrier or introduction of a new pathogen to a region that was previously free of disease. Several large outbreaks occurred in swine populations in Canada in the last decade. This includes porcine circovirus type 2 associated diseases, triple-reassortant variant of H3N2 influenza virus, followed by numerous other influenza virus variants. Clinical problems due to *Brachyspira hyodysenteriae* and *Brachyspira hamptonii* emerged in western Canada in late 2000s. The latest example of the emerging infectious diseases is the porcine epidemic diarrhea virus (PEDv) and the porcine delta coronavirus, both of which were introduced to Ontario in early 2014. Novel variants of porcine reproductive and respiratory syndrome virus (PRRSV) are also detected on a continuous basis.

Understanding transmission in populations is important for control strategies. Nonetheless, the structure of livestock industries is becoming increasingly complex and dominated by existence of multiple networks of pig flows and service providers. Biosecurity and management practices are becoming more standardized, and sometimes are considered jointly to form disease prevention strategies. This makes analysis within the framework of classical risk-factor epidemiology challenging. Veterinary practitioners who work with different species are skilled in dealing with protocols aimed at prevention of introduction, reduction of spread within populations, or even elimination of infection from different sites. For these reasons, many populations are continuously under some type of infection control practices. In addition, the focus of such strategies is frequently on the entire production systems. This requires novel approaches toward studying transmission of infectious pathogens in swine populations.

The starting point for implementation of effective control of infectious agents is based on accurate understanding of the within-herd dynamics. For many swine pathogens, such knowledge is extrapolated from experimental studies. Field studies and thorough field and outbreak investigations specifically aimed to investigate transmission are still infrequent in the scientific literature. Nonetheless, results from such investigations are important for complete understanding of disease under field conditions. Such investigations could lead to design of better infection control strategies. Therefore, the objective of this paper will be to discuss approaches to study within-herd transmission of infectious agents and discuss insight that could be gained from such additional analysis. The emphasis will be on influenza virus. Influenza is frequently considered as a disease with relative simple natural history. Yet, prevalence of exposure to this pathogen is high. It endemically circulates in many herds and in some systems it is causing considerable production issues, alone or in combination with other respiratory and systemic pathogens.

Spatial and temporal patterns for porcine reproductive and respiratory syndrome in Ontario swine herds

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Background

Porcine reproductive and respiratory syndrome (**PRRS**) is the most costly and complex disease affecting swine herds in North America. The PRRS Area Regional Control and Elimination (**ARC&E**) projects began in Ontario in 2010 with the aim of monitoring and aiding in the development of control/ elimination strategies, as well as decreasing the pressure of infection in different regions. The objectives of this study were to characterize swine herds participating in three different PRRS ARC&E Ontario projects, investigate spatial dependence and clusters of positive herds; and describe temporal patterns of PRRS in the region of Niagara.

Methodology

The study included projects located in three regions of Ontario: Niagara, Watford and Perth. Demographics and biosecurity data were obtained using a standardized questionnaire; sampling was conducted by herd veterinarians and included blood and/or oral fluids (9-11 samples per herd). All samples were processed in the Animal Health Laboratory (University of Guelph) and included ELISA and PCR. A herd was considered positive if at least one of the samples was positive after ruling out false positives.

Descriptive analysis was conducted using SAS v.9.3 and ArcGIS 10.1, clustering analysis was conducted using the D-function on R v.2.15.0; and cluster analysis was conducted using R v.2.15.0 and SaTScan v.9.1.1.

Temporal description was conducted using Stata-IC 10.

Results

A total of 75, 72, and 223 sites from the regions of Niagara, Watford, and Perth, respectively, were enrolled. Most herds for all regions were finishers (>46%); more than 40% of swine herds reported having a shower-in facility in place and from the herds that did not have it; the majority reported having at least *Danish entry*. The majority of herds from the three regions reported using continuous flow and external trucks for pig movement. The mean PRRS prevalence in swine herds at the end of 2013 was 16.7% and 48.2% for the regions of Niagara and Watford; and 40.6% for Perth at the beginning of 2014.

Spatial analysis showed no evidence of spatial clustering of PRRS-positive herds for any of the regions. Risk maps showed that herds located at the eastern region of Niagara, south western region of Watford and south central region of Perth are at a higher risk to be positive for PRRS compared to the other areas. Clusters were found in all regions using the scan statistic and spatial relative risk methods. For the region of Niagara, the prevalence of PRRS appeared to be decreasing over the years 2010-2013.

Conclusions

Areas where swine herds are at higher risk for being positive for PRRS virus were identified in the current study and could be used for surveillance and disease control purposes. Further elucidation is needed on the role of different networks on PRRS status in Ontario swine herds.

Acknowledgements

Ontario Ministry of Agriculture and Food and Ontario Ministry of Rural Affairs, Ontario Swine Health Advisory Board, Canadian Swine Health Board, Ontario Pork, Natural Sciences and Engineering Research Council, Agricultural Adaptation Council, Animal Health Laboratory (University of Guelph).

Development of Novel Feed Industry Fibre Degradation Enzymes

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Background and Implications

- Global annual production of biomass, primarily plant cell wall materials including feedstock and fibre feeds, is estimated at about 1.5 – 2 trillion dry tons with Canada's share at about 500 million dry tons (Narendra R. and Y. Yang, 2005, *Trends in Biotechnology* 23:22-27).
- Lignocellulosic feedstock pre-treatment (27%) and cellulase enzyme (16%) expenses represent about 43% of the total cellulosic ethanol production cost, which needs to be considerably reduced in order to produce more competitive cellulosic ethanol cheaper than corn ethanol.
- With application of effective fibre degradation enzymes, efficiency of dietary energy and carbon utilization can be improved to reduce grains (corn, wheat, barley, oats, rye and sorghum) annually used at about 1 billion tons as feeds for animal production on the globe scale.

Structural Recalcitrance of Plant Cell Wall Biomass

- Cell wall biomass recalcitrance refers to the natural resistance of cell wall materials to enzymatic deconstruction by limiting enzyme accessibility to microfibrillar internal surface areas. Cell wall materials are porous and dehydration of biomass materials at high temperature (~150°C) causes irreversible collapse of pores.
- Biomass materials consist of three structural units of microfibrils with lignin crosslinks, microfibrils surrounded by hemicelluloses and elementary fibrils with crystalline cellulose chain bundles further linked by intrachain and interchain hydrogen bonds and van der Waals forces.

Current Status of Commercial Fibre Degradation Enzymes

- Current commercial fibre degradation enzymes are largely free cellulases produced by genetically modified fungi such as *Trichoderma species*, *Humicola insolens* and *Aspergillus niger*.
- These enzymes are bi-modular and less effective in penetrating into microfibrillar internal surface areas in raw biomass materials. However, these enzymes are effective in processing pretreated biomass materials in light industries such as textile, paper and wood pulp polishing.
- Global industrial enzymes, as biocatalyst by-products, are annually valued at \$3 billion with half of the share for alpha-amylases used in processing grain ethanol. Phytases are the single largest feed enzyme valued at about \$700 million per year. Beta-Glucanases are another effective feed enzyme.
- Efficacy of cellulases and other hemicellulases is less consistent and their market value shares are relatively low. There is a scarcity of industrial lignin degradation enzymes available on markets.

Development of Novel Industrial “Designer Fibre Degradation Enzymes”

- Novel industrial “Designer Fibre Degradation Enzymes” need to be developed with the feature of single modular, low molecular weight or size and are highly penetrating into raw biomass microfibrillar internal surface areas for effective hydrolysis.
- Novel “Designer Fibre Degradation Enzymes” also need to degrade lignin, have specific and high hemicellulase activities and show processive high activity in breaking-down crystalline celluloses.
- Functional metagenomic library based screening has emerged to become a powerful research tool for the discovery and development of a new generation of the industrial “Designer Fibre Degradation Enzymes” from rapid turnover symbiotic microbiota of animals' gut systems.

Take Home Messages

- Novel “Designer Fibre Degradation Enzymes” need to be developed from the symbiotic microbiota of animals' gut for industrial applications by using functional metagenomic library research tools.

Financial support is from the OMAF-University of Guelph Gryphon's LAIRR Program.

FEEDING THE SOW: COMPARISON OF GESTATIONAL AND LACTATION PROGRAMS

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BACKGROUND – This trial was set up to be side-by-side comparisons of gestating and nursing sow feeding systems. Comparisons are made on sow performance (weight and condition changes and reproductive data) and piglet performance (growth rate and litter weight variability to weaning).

METHODOLOGY – A 2 x 3 factorial design was set up to compare two gestational and three nursing feeding programs. Gestational feeding systems were: “Conventional” – feed adjusted based on visual appraisal of producer, and “Kansas” – feed adjusted based on sow backfat and weight measurements. Nursing feeding systems were: “Conventional” – low feed offered to about day 3 and quickly ramped up to full feed, “Ramp” – on day 1, sows received 2.0 kg of feed, on days 2 and 3 sows received 3.0 kg of feed. From days 4 to 8 sows were ramped up as quickly as possible to 1.8 kg + 0.6 kg/piglet of feed. When this target level was attained, it was maintained until day 12. From day 13 onwards, the sow was fed *ad libitum*, and “Ad lib” – self-feeders where the sow could decide for herself how much she would like to consume from day 1 to weaning. Sow and piglet productivity were measured over three parities. Of the 316 sows that commenced the trial, 44 completed just one parity, 72 completed only two parities and 200 completed all three parities.

RESULTS – The data tables from this trial are too large to fit into this abstract – please send request for final report to email above if you wish a copy of it. Some interesting results were that the Kansas system resulted in increased feed offered (and cost) compared to the Conventional gestational feeding system without any improvement in reproductive performance. As well, the ad lib system of nursing feeding resulted in larger birth litter sizes (differences in weaned litter sizes were lost by cross-fostering), smaller (but more uniform) piglets.

BENEFITS TO THE SWINE INDUSTRY – There was not one system that could be declared with confidence to be the “best” system. However, each system has strengths that producers should be aware of and could consider given his/her own production system challenges.

ACKNOWLEDGEMENTS – We would like to acknowledge funding from the Ontario Ministry of Agriculture and Food, the Ontario Ministry of Rural Affairs and Ontario Pork for this project. We would also like to acknowledge the cooperation and dedication of the owners and employees of Bartside Farms where the trial took place. George Woudenberg was very helpful in setting up the ad libitum feeders and helping with data collection when necessary. Sarah Cook was helpful in helping to sort the data.

Prediction of Boar Taint in Major Breeds of Canadian Pigs Using Genetic Markers

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BACKGROUND: Alternatives to castration for reducing boar taint levels in pork are being sought, in part due to concerns for piglet welfare and market demands. One option is to reduce boar taint by using genetic markers associated with the synthesis and degradation of main compounds responsible for boar taint in genetic selection programs. The objective of this study was to investigate the feasibility of predicting the amount of androstenone and skatole in fat tissues of intact males using single nucleotide polymorphism (SNP) markers in candidate genes.

METHODOLOGY: A total of 3,474 pigs were genotyped for 97 SNP markers previously identified to be associated with boar taint. Taint measures of boars with slaughter weights between 90 kg and 150 kg and less than 300 days of age were used in analyses. Consumer acceptance levels of 1,000 ng/g for androstenone and 200 ng/g for skatole were considered. Since distributions of both compounds were highly skewed, the natural logarithms of androstenone and skatole were used as phenotypes. A total of 644 Duroc, 837 Landrace and 871 Yorkshire boars were used to test the ability of SNP markers to predict boar taint levels. A two-step analysis was performed using the SAS GLM procedure to adjust phenotypes for season, age and weight at time of sampling followed by marker assisted genetic prediction using genomic breeding value estimator (gebv) software. The oldest 80% of boars were assigned to a training group to use their markers and boar taint measures to predict boar taint genetic values for the youngest 20% of boars in a validation group.

RESULTS: A total number of 61, 80 and 83 of genotyped SNPs had a minor allele frequency (MAF) of greater than 0.05 in Duroc, Landrace and Yorkshire pigs, respectively. Approximately 47% and 5% of Duroc, 28% and 14% of Landrace, and 25% and 10% of Yorkshire pigs had androstenone and skatole levels above consumer acceptance thresholds, respectively. Correlations of the marker assisted genetic values with the adjusted values of androstenone and skatole levels in the validation groups were 0.35 and 0.05 in Duroc, 0.26 and 0.26 in Landrace and 0.40 and -0.05 in Yorkshire, respectively. The number of unfavorable alleles of SNPs associated with androstenone in Duroc boars in the validation group was significantly correlated ($r= 0.33$, $p<0.001$) with androstenone levels in fat. The percentage of animals with androstenone levels above the threshold were 20% and 76% for groups of animals with 10 and 15 or more unfavourable SNP alleles, respectively. The low correlations observed for skatole are likely a consequence of the lower incidence of values above consumer acceptance thresholds for this compound in the breeds.

BENEFITS TO THE SWINE INDUSTRY: These results show the potential of the markers as a tool to select breeding animals against high levels of androstenone, the compound most responsible for unacceptable levels of taint in this study.

ACKNOWLEDGEMENT: Funding was provided by Agricultural Adaptation Council of Ontario. Financial support was also provided by regional swine improvement centers across Canada and participating Canadian breeders.

Bacteriophage Based Control of *Salmonella* through the Pork Chain: Current Status and Future Trends

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Salmonella continues to represent a significant food safety issue in pork production and processing. Despite attempts to reduce the carriage of *Salmonella* associated with pigs and pork products the prevalence of the enteric pathogen remains high. Moreover, the increased incidence of multi-drug resistant strains along with virulence is a major concern to the extent that lobby groups are seeking zero-tolerance status for the Top 4 serotypes. Therefore, there is strong motivation to seek interventions with the pork chain to control *Salmonella*.

Bacteriophages have a long history of use in treating infections and are widely applied in Eastern Europe as an alternative to antibiotics. In principle bacteriophages are the ideal antimicrobial given that they specifically target pathogens of interest, self-replicating & self-limiting and non-toxic. Yet, applying bacteriophages to control pathogens in animals and on foods remains a challenge.

The presentation will provide a state-of-the-art with respect to the application of bacteriophages to control *Salmonella* in pigs during production, within the production environment, during processing and pork products. The results presented will be those generated by our group and from the work of others.

Bacteriophages used to reduce *Salmonella* during pork production has been met with moderate success in that the use of the biocontrol agent reduce prevalence of the enteric pathogen but did not result in complete elimination. In studies performed to date it has been shown that *Salmonella* levels initially decrease with the introduction of phages but then subsequently increase due to neutralization of bacteriophages by immune system. Yet, no evidence of phage resistance has been observed although a shift in the serotypes making up the *Salmonella* population can potentially occur. Bacteriophages have shown limited ability to control *Salmonella* in the pig environment given that an equilibrium between phages and host occurs. More success has been met in applying bacteriophages just prior to processing especially if applied to the skin at high levels (multiplicity of infection = 10). In one report, phages applied at an MOI = 10 supported a 4 log reduction of *Salmonella* thereby minimizing levels of the pathogen entering the processing line.

Bacteriophages applied to processed pork products do not undergo replication but rather inactivate pathogens in a process termed inundation or lysis from without. Here, multiple phages attach to the *Salmonella* host thereby destabilizing the membrane. As a consequence, *Salmonella* levels are initially decreased but the surviving populations initiate growth if held at temperature abuse conditions.

From studies performed to date it can be concluded that bacteriophages are most effective at decreasing the carriage of *Salmonella* associated with pigs just prior to processing. However, limited *Salmonella* control by phages is observed when applied within the production phase or on processed meat. In terms of future trends, one can envisage the application of bacteriophages in combination with other hurdles such as antagonistic bacteria or additional hurdles. The use of phage endolysins for pathogen control is another area of active research although has yet to be applied against *Salmonella* associated with pigs.

Porcine Epidemic Diarrhea Update

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One of the crucial functions of veterinary diagnostic laboratories such as Animal Health Laboratory (AHL) is to rapidly detect emerging animal diseases. The rapid spread of porcine epidemic diarrhea virus (PEDV) in the US during 2013 was a warning that an incursion of PEDV into Canada was likely just a matter of time. Hence, in July 2013 the AHL developed a test to rapidly detect PEDV in porcine samples and put in place a triplex porcine coronavirus PCR test to detect and differentiate PEDV from TGEV and PRCV, both of which are endemic in Ontario. The index case of PED in Ontario was on January 22, 2014. Four suckling piglets with diarrhea from a 500 sow farrow-to-finish operation were submitted to the AHL tested positive for PEDV. The mortality and morbidity in suckling piglets was 100%. Positive samples were also rushed to NCFAD, Winnipeg for additional testing and were confirmed as PEDV-positive on Jan 23. As of April 30, 58 clinical cases were confirmed in Ontario.

Subsequent to the discovery of porcine deltacoronavirus (PDCoV) in the US in February, 2014, PDCoV was detected by PCR in Ontario in March, 2014 – 6 sites are confirmed positive. Excellent support and collaboration among AHL, American and CFIA laboratories has allowed early detection of PEDV and PDCoV, and hence an early response to limit the impact of these emerging pathogens.

Zinc usage in pig production: can it cause antibiotic resistance?

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Background: Therapeutic doses of zinc ($\geq 2,500$ ppm) are commonly used as a prophylactic in nursery pig ration to prevent post-weaning *E. coli* diarrhea. However, recent research has identified a zinc resistance gene (*czrC*) which is co-located with other antibiotic-resistance genes in *Staphylococcus aureus* and *Staphylococcus hyicus*. Since these bacteria are widely disseminated in pigs, there is concern that exposure to zinc may be able to select for antibiotic-resistance in these bacteria in the absence of antibiotics. Selection of antibiotic-resistance in *S. aureus* and *S. hyicus* is unwelcome as these bacteria are common pathogens in humans and animals. The following study examined the effects of therapeutic zinc oxide (ZnO) on methicillin-resistant *S. aureus* (MRSA) in nursery pigs raised without antibiotics.

Methodology: An on-farm randomized-controlled trial was completed using 110 pigs from 23 sows from a herd naturally colonized with *czrC*-positive MRSA. Pigs were followed from birth to 49 d and sampled weekly. At weaning (21 d), stratified randomization was used to equally distribute MRSA-positive and MRSA-negative pigs into 8 pens; 4 pens received a control ration containing 100 ppm ZnO (minimum dietary levels) and 4 pens received a therapeutic ration containing 3,000 ppm ZnO. No other antimicrobials were used. Collected nasal swabs were tested for MRSA using an enrichment culturing method. Multivariate logistic regression was used to analyze the data and adjust for co-variate effects.

Results: Pre-weaning mortality and culling was 10% (11/110) but there were no losses after weaning. In total, 49 pigs received the control ration and 50 pigs received the therapeutic ration. MRSA was detected in 5 pigs (4.8%) within 24 hours after birth and persisted at a mean prevalence of 5.6% among suckling pigs. Prevalence of MRSA among sows was 35% (8/23). Following weaning, the prevalence of MRSA increased dramatically and was significantly higher in pigs fed 3,000 ppm ZnO at 1-week ($P=0.007$) and 2-weeks post-weaning ($P=0.015$) compared to controls. Persistence of MRSA (testing positive at ≥ 2 time-points) was observed in 2% (1/49) of control pigs and 22% (11/50) of treated pigs ($P=0.004$). At the end of the study MRSA was still detected in the group fed a ration containing 3,000 ppm ZnO but not among the control group, however this difference was not statistically significant ($P>0.05$).

Benefits to the swine industry: This study shows that antibiotic-resistance can be influenced by zinc in the absence of antibiotics; this may explain the presence and persistence of antibiotic-resistant staphylococci on antibiotic-free farms. Knowledge of in-feed zinc content is important for swine practitioners who may be treating antibiotic-resistant infections, including greasy pig disease which is caused by *S. hyicus*.

Acknowledgements: We thank OMAF/MRA and Ontario Pork for funding this research.

Whole body nitrogen retention in response to energy intake – testing the NRC 2012 model

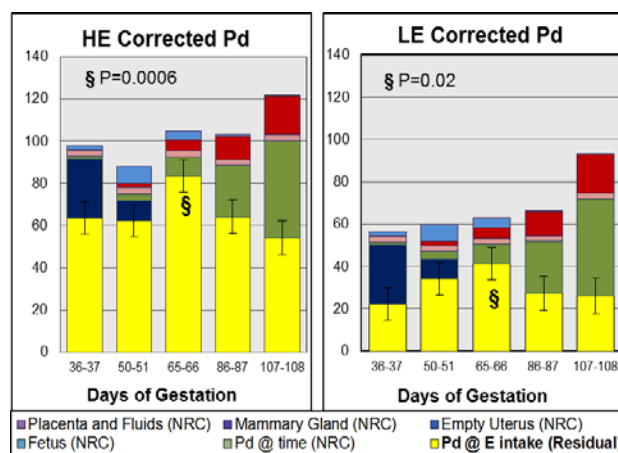
E.G. Miller, D.C. Wey, C.L. Levesque, and C.F.M. de Lange*

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Background: Whole body nitrogen (N) retention is a main determinant of amino acid (AA) and energy (E) requirements of gestating sows. However, information addressing the effect of E intake on maternal N retention during gestation is sparse (NRC, 2012). The relationship between maternal N retention and E intake (**Pd@E intake**) is assumed to be linear, constant throughout gestation, and to decline with increasing parity. More research is needed to characterize patterns of N retention, especially in parity 1 and 2 sows, where Pd@E intake is relatively large.

Methodology: 20 gestating purebred Yorkshire gilts at the University of Guelph Swine Research Station were selected at d30 of gestation (mean BW 166±3.5 kg; P2 backfat (BF) 20.7 ±0.84 mm) and fed a corn and soybean meal based diet (3300 kcal/kg ME, 17.8% CP), either at high (HE) or low (LE) energy level (NRC +15% or -15%, respectively). Maternal N retention was determined during five 4d N-balance periods starting at d35, 49, 63, 85, and 106 of gestation. Observed gilt performance over the entire gestation period was matched with predicted performance using the NRC (2012) model, by adjusting maintenance E requirements and the ‘mean’ slope of Pd@E intake over the entire gestation. Pd (N retention x 6.25) in each protein pool was calculated with the NRC (2012) model; after correction for the systematic difference between observed N retention and whole body Pd (calculated based on BW and BF gain); except Pd@E intake during each of the N-balance periods, which was calculated as a residual.

Results: Energy level (P<0.001) and day of gestation (P=0.001) affected total Pd, but without an interaction (P=0.853). To match observed performance with NRC (2012), default maintenance E requirements were decreased and the ‘mean’ Pd@E intake was increased. Mean observed corrected maternal Pd@E intake throughout gestation for HE gilts was 66g/d for HE gilts and 30g/d for LE gilts, but at d 65-66, Pd@E intake was greater than these mean values (P<0.001 and P=0.02 for HE and LE, respectively).



Benefits to Swine Industry: Calibrating the NRC (2012) model for this population of sows required a reduction in maintenance E requirements and an increase in maternal N retention, resulting in more E for body lipid deposition and increased estimated requirements for lysine and other AA. Moreover, maternal Pd related to E intake appears highest during mid-gestation. Thus, evaluation of the model with different genotypes to more closely predict sow performance is warranted in the future. This model will help nutritionists better formulate rations to reduce excess nutrient excretion and reduce feed costs where possible.

Acknowledgements: Thanks to lab mates and technical staff. Financial support provided by Ontario Pork, Ontario Ministry of Agriculture and Food, Royal de Heus, and Ajinomoto-Heartland Lysine.

Influence of steeping DDGS on growth performance and digestive function in weanling pigs

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Background: The high ratio of insoluble:soluble fiber contained in DDGS is suggested to stimulate gut health and digestive function in weanling pigs; however the high total fiber content limits energy availability. Steeping, the process of soaking feedstuffs in water for extended periods of time, of DDGS can be utilized as a strategy for improving utilization of energy from the fiber fraction, particularly when mixed with exogenous carbohydrases. Combining innate benefits of feeding DDGS with steeping may improve energy utilization and digestive function in newly weaned pigs.

Methodology: Effect of feeding DDGS on digestive function and growth performance, and the influence of steeping DDGS were evaluated in newly weaned pigs (initial BW=6.02±2 kg, 14 pigs/pen, 6 pens/treatment) in two experiments. In experiment 1 pigs were fed dry corn and soybean meal based diets: a control or a diet containing DDGS at 7.5% in phase 1 (d 0-7), and 25% in phase 2 (d 7-20) and 3 (d 21-35). In experiment 2 pigs were liquid fed DDGS containing diets (as in experiment 1) with a β -glucanase and xylanase; (67.2 and 51.4 IU/g DDGS resp., AB Vista): unsteeped (usDDGS; diets mixed with water at feeding (28% DM)) or steeped (sDDGS; DDGS with enzymes steeped between d5 and 14 in 39°C water at 16% DM prior to feeding). For each treatment 3 pigs/pen were euthanized for evaluation of gut and digesta characteristics on d7, 14, and 35.

Results: Dietary treatment did not affect ($P>0.10$) overall (d 1-35) growth rate or feed efficiency in either experiment. In experiment 2 batch characteristics ($n=5$, mean±sd) of sDDGS (d5 and d14 resp.) revealed lactic acid 127.1 ±22.3 and 90.6 ± 26.6 mM, and acetic acid 68.0 ± 18.3 and 145.6 ±33.5 mM. Preliminary data on digesta characteristics indicate a treatment effect on gut health and digestive function. In both experiments dietary treatments influenced bacterial metabolite profiles throughout the digestive tract. More specifically, in experiment 1 control pigs had higher ($P<0.05$) d14 cecal n-butyric acid (32.4 vs. 22.3 mM) and tended to have higher jejunal lactic acid on d14 ($P=0.07$) and 35 ($P=0.08$) (102.3 vs. 49.0 and 111.0 vs. 60.7 mM). Dry DDGS fed pigs had higher ($P<0.05$) d35 ileal acetic acid (7.3 vs. 12.4 mM), higher ($P<0.05$) d14 jejunal and ileal propionic acid (9.8 vs. 29.1 and 14.5 vs. 35.2 mM), and tended ($P=0.07$) to have higher d35 cecal acetic acid (82.7 vs. 108.4 mM). In experiment 2 sDDGS pigs had higher ($P<0.05$) d14 jejunal n-butyric acid (42.1 vs. 61.8 mM) and tended ($P=0.09$) to have higher cecal lactic acid (23.7 vs. 48.6 mM) while usDDGS pigs tended ($P=0.08$) to have higher d35 ileal lactic acid (42.1 vs. 28.9 mM).

Benefits to the swine industry: Results indicate some potential for adding value to DDGS by utilizing and improving gut health and digestive function benefits during the vulnerable post-weaning period. There is a need to more tightly control liquid feed conditions and further explore bacterial and enzyme combinations to maximize benefit.

Acknowledgements: Funding was provided by OMAF, NSERC, and industrial partners of swine liquid feeding association (www.slfa.ca)

HOG HEART HEALTH AND IN TRANSIT LOSSES

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BACKGROUND- The increase in shipping mortalities during the summer months is often attributed to heat exhaustion or stress. In Ontario, approximately 0.07% of hogs shipped to slaughter die during transport to the plant. However it is rarely questioned why so many hogs are able to tolerate the shipping conditions and why only a small percentage cannot. Research from the authors (KZ, TVD) and other published studies have suggested that heart failure may be the cause of death for the majority of in-transit losses. However factors that contribute to why the hogs develop cardiomyopathies are not known. The goal of this project was to examine and characterize heart lesions in market hogs and test for a genetic association for the cardiomyopathies observed.

METHODOLOGY- From May 2012 to September 2013, hogs that died while in transit (InTransit Loss-ITL) to one slaughter plant were examined at the Animal Health Laboratory, University of Guelph. The hearts were removed from the carcasses and fixed in 10% formalin for further examination. Hearts were also collected from hogs that did not die in transit (non-ITL) from the slaughter line at the same abattoir. Heart weights for hogs that died in transit were compared to the hearts of hogs that did not die in transit. Each heart was examined by one veterinary pathologist (TVD) both visually and microscopically. Tissue samples from a selection of the hearts were sent to Geneseek Labs Inc. (Nebraska, USA) for gene sequencing. These sequence data were analyzed to determine if a common gene or genes could be associated with pigs that die during transport.

RESULTS- Post mortem findings on ITL hogs indicated cause of death was consistent with heart failure. Table 1 lists the average heart weights which were significantly heavier for ITL hogs.

Visible enlargement of the heart was more common in the hearts of hogs that died in transit. The ITL hearts had hypertrophy of the left and/or right ventricle at a rate of 93% (77/83). The non-ITL had hypertrophy of the left and/or right ventricle at a rate of 7% (5/67).

These pathologic changes were considered to have developed weeks to months prior to death.

38 ITL hearts and 34 non-ITL hearts had samples sent for genetic sequencing and analyses. Analyses of the gene sequencing data showed over 40 genes possibly associated with a pig dying during transport. Two of these genes are known to be associated with HCM in humans.

BENEFITS TO THE SWINE INDUSTRY- Early results from this study indicate that the majority of hogs that die in-transit to the abattoir die as a result of pre-existing heart lesions and suggest that these cardiomyopathies may have a genetic association. Further testing and analyses are needed to identify specific gene(s) involved. If this can be established, shipping mortalities could be reduced through genetic selection to eliminate a HCM-like gene or genes from market hogs.

ACKNOWLEDGEMENTS- Funding for this project was provided by an Ontario Pork Research Grant and by the Agricultural Adaptation Council’s Canadian Agricultural Adaptation Program.

Table 1. Average heart weights of ITL and non-ITL swine hearts

Heart type	Total Heart Weight (THW) [SD]	Left Ventricle + Septum (LV+S) [SD]	Right Ventricle (RV) [SD]	LV+S/ RV [SD]	THW/ body weight [SD]
ITL N=83	442.0 [±66.4]	275.0 [±43.7]	98.5 [±21.4]	2.9 [±0.5]	3.7 [±0.5]
Non-ITL N=67	368.8 [±37.9]	243.0 [±25.6]	83.0 [±13.3]	2.9 [±0.4]	3.1 [±0.3]
P value	0.0001	0.0001	0.0001	0.112	0.0001

Carcass and Meat Quality for Immunocastrated and Entire Males from Different Genotypes

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BACKGROUND- Immunocastration is an animal welfare friendly alternative to surgical castration which involves vaccination against gonadotrophin releasing factor. The vaccine (Improvac, Zoetis) is administered in 2 doses. The first dose primes the immune system of piglets for the second dose of Improvest without altering testicular size or function; the second dose is administered at 4~6 weeks prior to slaughter to stimulate the protective immune response inhibiting testicular function along with metabolizing and removal of boar taint compounds. Piglets can grow as an intact boar with greater growth rates and carcass leanness. Data are lacking in the industry for administering the 2nd dose earlier than 6 weeks preslaughter. Ractopamine is a commercially available β -agonist used to increase protein deposition and growth rate during the terminal stages of finishing to ultimately improve feed efficiency along with increasing lean meat yield. While past work has found that ractopamine can be used successfully with immunocastration, data are lacking for examining different pig genotypes, use of distillers dried grains plus solubles (DDGS) in finishing diets, and marketing pigs at the heavier weights currently found in North America. The objective of this study is to examine the interactive effects of pig genotype, immunological castration, and use of ractopamine on growth performance, carcass traits, and pork quality of entire male pigs fed diets containing DDGS.

METHODOLOGY- 146 entire male pigs representing 2 genotypes (PIC genetics from a high meat quality line vs. Large White genetics selected for growth). 5 management regimens based on gender (entire male vs. immunological castration using Improvest) and Terminal Finisher diet (Control vs. a ractopamine supplemented diet for the last 21 days on feed) were evaluated including: Entire males fed Control diet, Entire males fed ractopamine, Immunologically castrated males (fed Control diet) with 2nd dose administered 8 weeks pre-slaughter, Immunologically castrated males (fed Control diet) with 2nd dose administered 4 weeks pre-slaughter, Immunologically castrated males (fed ractopamine) with 2nd dose administered 4 weeks pre-slaughter. Pigs were slaughtered when average pen weight was 125 kg. Detailed evaluations of carcass cutout, meat quality, and fatty acid composition were conducted post-slaughter.

RESULTS- There were few management regimen effects (use of immunocastration with or without ractopamine) on growth performance traits and most measures of meat quality. Use of ractopamine produced heavier carcasses with lower fat deposition (backfat and total fats) and more muscling, and dissectible lean but at the expense of decreasing loin tenderness. Administration of the 2nd dose of Improvest at 8 vs. 4 weeks pre-slaughter decreased feed conversion and increased backfat deposition; these pigs were more like conventional barrows. There was generally no management regimen by breed interactions for the traits evaluated.

BENEFITS TO THE SWINE INDUSTRY- The current study provides useful data that earlier administration of the 2nd dose of Improvest results in performance of immunologically castrated males being more like conventional barrows. In addition, use of ractopamine was not beneficial from a growth performance standpoint when marketing heavy boars. There was generally no management regimen by breed interactions involving immunocastration.

ACKNOWLEDGEMENTS – We would like to thank Zoetis for providing Improvest for the trial. We sincerely appreciate the help of Doug Wey, Yanping Lou, and Ponsonby General Animal Facility and Meat Lab staff in the study.

THE EFFECTS OF ANOXIC STATIC GROWTH AND GROWTH PHASE ON THE EXPRESSION OF *ACTINOBACILLUS SUIIS* ADHESIN GENES

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BACKGROUND - *Actinobacillus suis* is a common commensal of tonsils of the soft palate of swine, but unknown stimuli can lead to invasive, systemic disease. Its pathogenesis, including the process of host colonization, is poorly understood. Thus, the objective was to measure expression of genes involved in attachment, a key factor in both health and disease.

In healthy animals, *A. suis* is thought to exist in tonsils in biofilm and planktonic forms. Cells in biofilm likely persist in a lower oxygen/nutrient environment in stationary phase. Cells shed from the biofilm assume a planktonic form and have higher nutrient/oxygen availability. We hypothesize that *A. suis* will differentially express adhesins in these environments, and that certain signals will lead to an invasive phenotype with a different complement of adhesins.

METHODOLOGY - From 42 adhesin-associated genes (23 adhesins) identified by bioinformatic analysis of the virulent *A. suis* H91-0380 strain, 9 genes were chosen for RT-qPCR. RNA was prepared from aerobic cultures grown at 37°C and 200 rpm and sampled at 60 min post-inoculation (mpi) and 180 mpi for exponential and stationary phase, respectively, and from anoxic static cultures grown at 37°C+5% CO₂ and 0 rpm and sampled at 60 and 210 mpi.

RESULTS - All genes were up-regulated in one growth phase: type IV pilin *ppdD*, outer membrane proteins *ompA2* and *ompP2*, and fibronectin-binding *ybaV* in exponential phase, while biofilm-associated *flp*, fine-tangled pili *ftpA*, filamentous hemagglutinin *fhaB*, *ompA1*, and autotransporter *tibA* were up-regulated in stationary phase. Most genes were up-regulated in one growth condition: *ftpA*, *ompA1*, and *tibA* in aerobic, and *flp*, *ppdD*, and *ompA2* in anoxic growth. Time by treatment interactions were also observed for several genes (Fig.1). Work is underway to generate knockout mutants of the adhesin genes to determine their roles in attachment and biofilm formation.

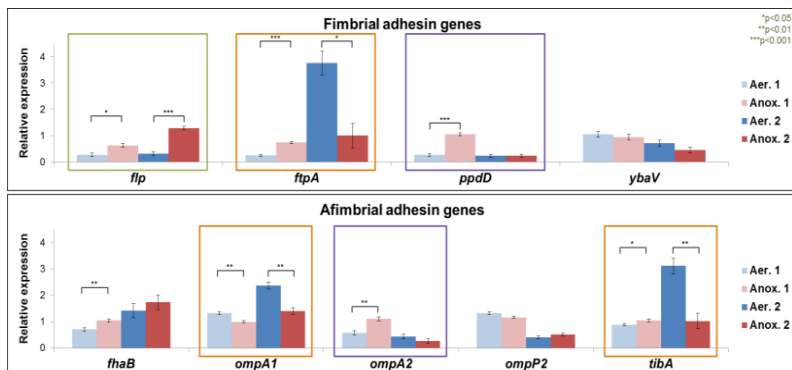


Fig. 1 Time*Treatment effect of *A. suis* adhesin gene expression during aerobic, anoxic static, exponential, and stationary growth.

BENEFITS TO THE SWINE INDUSTRY - This project should provide insight into the mechanism of invasion of *A. suis* and other systemic pathogens and could help in the creation of live attenuated and other vaccines.

ACKNOWLEDGEMENT – Funded by NSERC, OMAFRA, OGS and OVC.

Epidermal Growth Factor enhances Small Intestine Development in Early-Weaned Pigs: Potential *in vivo* Mechanisms Involved

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Epidermal growth factor (EGF) is one of the key growth factors in multiple species, including humans, cattle, and pigs. We have previously generated EGF expressing *Lactococcus lactis* (EGF-LL) and shown that feeding the supernatant of this fermentation to newly-weaned piglets improves growth performance. In this study, we sought to determine the mechanism behind the improved growth and intestine development stimulated by EGF. Sixty-four newly weaned pigs aged 19-21 days were assigned to one of two treatment groups: 1) supernatant of EGF-LL fermentation product, after removal of EGF-LL (SuperEGF), or 2) blank M17GE media (Control). Each treatment group consisted of 4 pens, with 8 pigs per pen. EGF-LL supernatant was administered for the first 3 weeks post-weaning. Consistent with our previous report, overall average daily gain (433.4 ± 10.9 vs 388.7 ± 7.8 g BW gain/day) and Gain:Feed (0.76 ± 0.03 vs 0.68 ± 0.01 kg gain/kg feed) of EGF treated pigs were significantly improved in comparison to control pigs ($P < 0.05$). Jejunal villi height was also increased by EGF supplementation (588.9 ± 28.2 vs. 511.4 ± 21.5 μm). As glucagon-like peptide-2 receptor (GLP2R) mediated signaling is a key factor in intestine development, we next studied the influence of EGF on the expression of GLP2R and its down- stream factors, keratinocyte growth factor (KGF), and sodium glucose cotransporter (SGLT1). It was revealed that the expression of GLP2R, KGF and SGLT1 were up regulated by EGF *in vivo*. Goblet cell number, a known KGF responsive intestine differentiation marker, mucin 2 production, and alkaline phosphatase activity were also increased, suggesting an enhanced differentiation by EGF. To assess the inflammation status of the early weaned pig intestine, intraepithelial lymphocytes, and villus lamina propria widths were examined. While no difference was observed in intraepithelial lymphocyte counts, villus lamina propria widths were significantly decreased by supplementation of EGF. Additionally, levels of jejunal IL-13, an anti-inflammatory cytokine, were significantly increased with EGF treatment, which might account for the decreased lamina propria width observed. Together, our data suggest that EGF increases intestine health, and development, and early weaned pig growth by influencing the expression of an array of key intestinal development genes, effecting multiple downstream pathways. A model outlining of the possible mechanisms behind EGF action in the small intestine in this important stage is proposed.

High-level expression of porcine protegrin-1 in *Pichia pastoris* for Potential Use as an Alternative to Conventional Antibiotics

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Antibiotics remain at the forefront for treatment of microbial infections in humans and animals; however, the increase prevalence of bacterial resistance to conventional antibiotics is a growing public health concern. To overcome antibiotic resistance, searching for effective antibiotic alternatives is a challenge. Protegrin-1 (PG-1) is a porcine cathelicidin antimicrobial peptide (AMP) that can exert its activity against a broad range of microorganisms including bacteria, fungi and enveloped viruses. PG-1 has been well studied and structurally characterized allowing it to be an attractive candidate for therapeutic use as an alternative to antibiotics. However, chemically synthesized AMPs are too costly to be feasible for routine use in the food-animal industry. Recombinant expression of PG-1 using a safe and nonpathogenic microbe such as yeast is a potentially inexpensive alternative approach to chemical peptide synthesis for larger-scale food and animal application. Here, we report a codon-optimized protein corresponding to proform PG-1 that is highly expressed in *Pichia pastoris* yeast. For potential oral application of recombinant proform PG-1, an intestinal enterokinase (EK) cleavage site was included for EK-mediated activation of the cathelicidin. A *P. pastoris* transformant harboring multiple copies of the expression cassette was selected for downstream expression studies. Secreted proform PG-1 reached 1.2 g/L at 52 h during high cell density bioreactor cultivation. The unpurified proform PG-1 exhibited high antimicrobial activity against *S.aureus* and *E.coli* after *in vitro* EK cleavage (minimum inhibitory concentration 10 and 2 µg/mL, respectively). Resulting data establishes the potential feasibility of using microbes, in particular the yeast *P. pastoris*, as bioreactors to express and secrete biologically active animal-derived AMPs for potential large-scale application.

Fermenting soybean meal to enhance digestibility and nutrient value using novel isolated microbes

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Animal-based proteins are common, but expensive sources of protein for piglet feed in the livestock industry. For this reason, and also due to the concern of potential pathogen spread by animal plasma, there is an increased interest in using plant-based proteins as a protein source in livestock feeds. Soybean meal continues to be a valuable source for protein in the adult animal. However, issues such as the anti-nutritional factors found in soybean and the immature gastrointestinal tracts (GIT) of piglets limit the use of soybean meal in the diets of young animals. These anti-nutritional factors, which include oligosaccharides, polysaccharides, and large, antigenic proteins, have negative effects on overall growth performance¹. Past studies using bacteria-fermented soybean meal have shown encouraging results for reducing these anti-nutritional factors, increasing amino acids, and improving piglet growth performance^{2,3}. The objective of the current study is to improve the fermentation of soybean meal by using a newly isolated strain of bacteria. Bacteria, isolated from fermented food sources, were used to ferment soybean meal in a solid-state condition. Fermented samples were lyophilized and used for peptide profiling and analysis of crude protein and amino acids. SDS-PAGE results demonstrated a decrease in the quantities of antigenic proteins and high-molecular weight peptides, while also showing an increase of low molecular weight peptides. Amounts of crude protein and total amino acids showed a significant increase after 24 hours using Isolate-2. Further investigations of the change in oligosaccharide and non-starch polysaccharide content when fermented with Isolate-2 are still ongoing. In conclusion, fermentation of soybean meal using our newly isolated strain can effectively improve soybean meal, making it a cost effective, highly digestible protein source for piglets.

Energy intake and nutrient digestibility in heavy finishing swine fed varying levels of soluble fiber

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Background: The use of different types of dietary fiber in swine nutrition is of interest. Potential effects of fiber are to depress daily energy intake and utilization of dietary nutrients in growing pigs. These effects are likely to decline with increasing body weight. This study was conducted to determine the effect of increasing dietary levels of soluble fiber on daily feed and energy intake, as well as apparent fecal nutrient and energy digestibility in finishing pigs.

Methodology: In total 36 individually housed barrows (commercial crossbred; initial BW 78.38 ± 0.32 kg), were assigned to 4 dietary treatments in a randomized block design: control (corn and soybean meal based; 3229 kcal/kg calculated ME, 13.9% CP, 1.5% soluble fiber) and three isonitrogenous diets with increasing levels of soluble fibers (4, 8 and 12%; from added pectin, containing 63% of soluble fiber), and consequently decreased levels of calculated ME (3095, 3017 and 2946 kcal/kg). Pigs were fed ad libitum. At a BW of 105 kg, feces were collected twice daily, during three days. Insoluble ash was used as marker for determining digestibility. Statistical analyses were performed using the PROC MIXED procedure of SAS.

Results: The results showed a linear decrease for both daily feed intake ($P = 0.0157$) and daily DE intake ($P = 0.0005$) between 100.86 and 126.19 kg BW. Quadratic effects ($P < 0.0001$) were observed for the digestibility of DM, GE and CP; the maximum digestibility values were obtained at 0.74, 1.33 and 1.60% additional soluble fiber in diet, respectively. There was linear decrease ($P < 0.0001$) for the digestibility of ash. These results confirm that feedstuffs containing soluble fiber can be used to reduce daily energy intake in heavy finishing pigs through qualitative feed restriction, but its negative effect on the nutrient digestibility must be considered.

Table 1. Daily feed (kg/d) and DE intake (kcal/d), as well as apparent fecal digestibility (AFD, %) of energy and nutrients in finishing pigs fed diets containing different levels of soluble fiber (SF).

	Experimental diets (% SF)				SEM	Effect of diet SF
	1.5	4	8	12		
Daily feed intake	1.64	1.56	1.61	1.34	0.18	Linear
Daily DE intake	6822	5792	5937	4925	317	Linear
AFD						
DM	89.75	89.26	87.54	85.35	0.32	Quadratic
GE	89.92	90.19	88.21	86.29	0.33	Quadratic
Ash	51.91	45.38	34.94	28.05	1.09	Linear
CP	86.64	87.63	84.99	82.90	0.69	Quadratic

Benefits to the swine industry: Knowledge about soluble fiber digestibility and its effects in swine nutrition could be useful to increase profits in the swine production, making the use of co-products from food and bio-product industries viable.

Acknowledgements: Financial support was obtained from FAPESP (Foundation for the Support of Research of the State of São Paulo) through the process 2012/10206-2

Increased energy intake affects maternal nutrient reserve and indicators of insulin sensitivity in gilts during late gestation

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Background: During late gestation, energy requirements for fetal growth increase rapidly, which causes increased glucose shunting to the fetus. This may be facilitated through the development of insulin resistance in the sow, mediated in part through increases in cortisol levels¹, and resulting in maternal lipid mobilization². Thus, increasing dietary energy intake during late gestation may prevent a negative energy balance and improve sow reproductive performance². However, insulin resistance may have consequences for energy utilization, including voluntary feed intake during lactation. This experiment was conducted to evaluate the effect of energy intake during gestation on sow energy status in late gestation (i.e. extent of body lipid mobilization and blood levels of ketones [eg. β -hydroxybutyrate, BHB] and non-esterified fatty acids [NEFA]).

Methodology: 52 gestating purebred Yorkshire gilts at the University of Guelph Swine Research Station were assigned to a high (HE) or low (LE) energy level (NRC +15% or - 15%, respectively) upon pregnancy confirmation at day 30 of gestation (mean BW 168±2.0 kg; P2 backfat (BF) 20.2 ±0.5 mm). A corn and soybean meal-based diet was used for both energy levels (3300 kcal/kg ME, 17.8% CP). At days 82 and 110 of gestation, blood samples were taken from suborbital sinus for analysis of NEFA, BHB and cortisol. BW and BF were measured at day 67, 82, 92 & 110 to estimate average daily gain (ADG) and total body lipid (BL) and protein content (BP)³.

Results (Table 1): Serum BHB in HE gilts was greater in late gestation (P=0.0004). Higher energy during gestation increased BF and BL gain, which may have resulted in an insulin resistant state and decreased glucose use in peripheral tissues during late gestation. Serum NEFA levels did not differ between HE and LE gilts (P=0.016). Serum cortisol was also not affected by energy intake (P=0.19).

Table 1. Effect of treatment (high or low dietary energy, mean ± SE) on serum BHB, NEFA & cortisol (d82 & 110), and body composition characteristics in gestating gilts (d67-110)

	High	Low	P value
BHB (umol/L), n=103	13.5 ± 1.3	6.87 ± 1.3	0.0004
NEFA (mmol/L), n=103	0.077 ± 0.0061	0.098 ± 0.0058	0.016
Cortisol (µg/dL), n=83	22.0 ± 1.3	19.5 ± 1.4	0.19
Body Composition, n=52			
Mean BW (kg)	210 ± 1.1	194 ± 1.1	<0.0001
Mean P2 BF (mm)	19 ± 0.27	17 ± 0.26	<0.0001
ADG BW (kg/day)	0.75 ± 0.029	0.52 ± 0.029	<0.0001
ADG BL (kg/day)	0.18 ± 0.017	0.088 ± 0.017	0.0006
ADG BP (kg/day)	0.12 ± 0.0069	0.094 ± 0.0068	0.003

Benefits to the Swine Industry: Increasing dietary energy intake by 30% increased ketone production during late gestation in gilts. Increased blood ketone levels may be indicative of increased insulin resistance during late gestation, which has been shown to reduce voluntary feed intake until mid-lactation⁴. This experiment demonstrated that the optimum energy intake level during late gestation reflects a balance between meeting both the maternal requirements and the increasing demands of the fetus, in order to avoid excessive lipolysis.

Acknowledgements: Thanks to lab mates and technical staff. Financial support was provided by Ontario Pork, Ontario Ministry of Agriculture and Food, Royal de Heus, and Ajinimoto-Heartland Lysine.

¹Ryan *et al.* (1998); ²Père *et al.* (2000); ³NRC (2012); ⁴Weldon *et al.* (1994)

Whole body nitrogen retention is transiently improved in young pigs supplemented with excess leucine during endotoxemia

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Background: Inflammation causes a rapid increase in circulating pro-inflammatory cytokines that coordinate an acute phase response, limiting whole body nitrogen (N) retention (i.e. lean growth) and disrupting normal N and amino acid (AA) metabolism. Leucine (Leu) is able to regulate muscle protein metabolism independently of growth factors and hormones and may act to modulate whole body protein metabolism in the pig during inflammation. The objective of this study was to explore the effect of dietary leucine supplementation on the dynamics of whole body N retention in starter pigs after an endotoxin challenge.

Methodology: Yorkshire barrows (two blocks of 12 pigs per block, initial BW = 13.9 ± 2.0 kg) were assigned to one of four treatments (2 × 2 factorial, n = 6). Pigs were fed isoenergetic and isonitrogenous diets that contained Leu 10% above requirements for protein deposition (CON; 1.36% SID Leu) or Leu supplemented at twice that amount (+LEU; 2.72% SID Leu). Immune system stimulation (ISS) was either induced with repeated intramuscular injections of bacterial lipopolysaccharide (ISS+; initial LPS injection = 30 µg/kg BW, increased by 20% with each consecutive LPS injection on days 3, 5, and 7) or not induced with repeated injections of saline (ISS-). The pigs were adapted to experimental diets for 5 d and then adapted to metabolism crates for 2 d. The pigs were fed restricted amounts of the experimental diets every 4 h according to BW. Whole body N retention was measured daily using standard N balance procedures. Blood was collected on day 1 pre-challenge and day 7 post-challenge 1 h after feeding for plasma albumin (Alb), haptoglobin (Hapt), and AA concentrations.

Results: Pigs fed +LEU had higher plasma Leu than pigs fed CON (319 vs 159 µmol/L, SE 10.3, P < 0.01). There were no differences in Alb and Hapt between ISS- and ISS+ pigs on day 1 pre-challenge (P > 0.10). On day 7 post-challenge, ISS+ pigs had lower Alb and higher Hapt than ISS- pigs, indicating that LPS-induced ISS was successful. There was no effect of diet on whole body N retention across the 7-d post-challenge N balance period (P > 0.10). However, ISS increased urinary N excretion and decreased whole body N retention during this period (P < 0.05). On day 2 post-challenge, whole body N retention was (1) lower in ISS+ pigs fed CON than ISS+ pigs fed +LEU (10.48 vs 12.28 g/d, SE 0.66, P < 0.05); (2) higher in ISS- pigs fed CON than ISS+ pigs fed CON (13.03 vs 10.48 g/d, SE 0.66, P < 0.05); and (3) not different between ISS- and ISS+ pigs fed +LEU (12.33 vs 12.28 g/d, SE 0.66, P > 0.10).

Conclusions and implications: Inflammation compromises the efficiency of AA utilization for whole body N retention in pigs. In this study, LPS-induced ISS transiently decreased whole body N retention in pigs and dietary Leu supplementation partly attenuated the subsequent reduction in whole body N retention. The mechanism by which Leu improves whole body N retention after an LPS challenge should be further explored.

Acknowledgements: Financial support was provided by Ontario Pork, the Ontario Ministry of Agriculture and Food, and Swine Innovation Porc.

Urea-nitrogen has lower bioavailability than ammonia-nitrogen for improving growth performance of pigs fed nitrogen-limiting diets

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Introduction: Non-protein nitrogen (NPN) absorbed in the lower gut (i.e., caecum and large intestine), primarily in the form of ammonia, can be used for improving nitrogen (N) retention and body weight (BW) gain in pigs fed a diet deficient in non-essential amino acid (NEAA)-N (Mansilla et al., 2013). In addition, supplementing NPN (ammonium salts) in diets deficient in NEAA-N can improve feed efficiency in growing pigs. Moreover, we have shown that NPN is as efficient as intact protein (i.e., casein) for providing dietary N for endogenous synthesis of NEAA (Mansilla, 2013). Although these studies indicate that NPN can be effective in low-protein pig diets, further understanding of N utilization and metabolism is warranted.

The objective of the present study was to determine the efficiency of using different dietary sources of N (NPN and specific NEAA) for improving growth performance of pigs fed diets limiting in NEAA-N.

Methodology: In total, 72 Yorkshire barrows with an initial BW of 16 kg were housed in individual pens. Pigs were randomly assigned to 9 different diets and growth performance was monitored during 3 consecutive weeks. Pigs were fed restricted at 3.0 x maintenance energy requirements (NRC, 2012). The basal diet contained casein and crystalline essential amino acids (EAA) as the only N sources and exceeded requirements for all EAA of a growing pig (NRC, 2012), but was low in crude protein (CP, N x 6.25, content 8.0 %). Different sources of N (urea, ammonia, glutamate and a mix of NEAA) were added to the basal diet to provide extra 1.35 and 2.70 % CP. The added mix of NEAA was based on the NEAA profile of body protein and was anticipated to minimize the need for endogenous NEAA synthesis. Blood samples were taken at the end of the trial to determine plasma urea-N concentration.

Results: Body weight gain and feed efficiency were similar ($P > 0.05$) and increased linearly with level of supplemented ammonia, glutamate and NEAA mix ($P < 0.05$; mean values for BW gain were 380, 421, 479 g/d for 3 respective levels of CP; average across these N sources) but not with urea ($P > 0.05$; 380, 381, 394 g/d for 3 urea levels). Urea-N plasma concentration was increased only in pigs fed urea ($P < 0.05$; 2.42, 11.6, 19.2 µg/ml) but remained low and similar when feeding ammonia, glutamate and NEAA mix ($P > 0.05$; mean values 2.42, 2.68, 3.76 µg/ml).

Conclusions and implications: Ammonia-N is as bioavailable as glutamate or NEAA mix-N for increasing BW gain and feed efficiency of pigs fed a diet limiting in NEAA-N. Urea-N has lower bioavailability than ammonia-N for improving pig growth performance. Dietary CP or NEAA-N should be carefully considered when diets are formulated with large amounts of crystalline EAA.

Acknowledgement: Financial support was provided by Evonik Industries, Ontario Pork, OMAF and Swine Innovation Porc.

INVESTIGATION OF THE USE OF KETOPROFEN ON DYSTOCIC SOWS POST-FARROWING FOR IMPROVING PIGLET PERFORMANCE

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BACKGROUND

Parturition is a painful process, particularly in the case of dystocia. The alleviation of pain in the nursing sow is likely to improve animal welfare and may impact growth performance in the suckling piglets. Non-steroidal antiinflammatory drugs (NSAID) may prove efficacious in the treatment of postparturient disorders and pain associated with a dystocic parturition. The objective of the study was to determine the effects of administering ketoprofen to dystocic sows shortly after parturition on maternal health, nursing behaviour, and piglet survival and growth.

METHODOLOGY

The study involved 3 farms and included 16 sows and their litters (196 piglets). Dystocic sows were systematically allocated to receive a single IM injection of 3 mg/kg of bodyweight of ketoprofen (Anafen®, Merial Canada Inc., Baie D'Urfé, QC; n=8) or a similar volume of a placebo (n=8) within 20 min of farrowing completion. Blood ketone concentration and rectal temperature of sows were measured at treatment and 24 h later, and rectal temperature and feed intake were measured for 3 days. Mortality and growth rate were monitored for all piglets in the study, in addition to blood glucose concentration and rectal temperature measured within 90 min of birth and 24 h after birth. Descriptive statistics and quantitative statistical analysis were completed in STATA 10_{MP} (Stata Corp., College Station, Texas, USA). Mixed-effect linear regression models were run to test for significant effects and interaction.

RESULTS

Average daily gain (ADG) and blood glucose level of piglets from sows treated with ketoprofen compared to piglets from sows receiving placebo is shown in Table 1. Blood glucose level at 24 h after birth was found to be positively correlated with average daily gain in the preweaning period ($P<0.001$). Feed intake of sows at 24 h after treatment was negatively correlated to rectal temperature at 24 h and 72 h ($\rho = -0.5$, $P=0.04$). Sows with a fever (rectal temperature, $\geq 39.5^{\circ}\text{C}$) at 24 h post-treatment consumed less feed than sows not having fever ($P=0.03$).

Table 1: Mean (SD) ADG and blood glucose concentration of piglets from sows treated with ketoprofen (treatment 1) or placebo (treatment 2) following a dystocic farrowing.

Time after birth	Treatment 1	N	Treatment 2	N	P value
	ADG (kg/day)				
0-24 h	0.133 (0.12)	92	0.061 (0.12)	92	0.004
0-48 h	0.179 (0.16)	88	0.086 (0.12)	90	<0.001
0-72 h	0.160 (0.11)	85	0.142 (0.05)	90	0.02
	Blood glucose concentration (mmol/L)				
<90 min	3.53 (1.8)	98	3.23 (1.6)	88	0.27
24 h	6.21 (1.6)	92	5.33 (1.4)	93	<0.001

BENEFITS TO THE SWINE INDUSTRY

This research provides evidence that the use of ketoprofen after dystocic parturitions may prove useful in the treatment of post-parturient disorders resulting in improved piglet performance.

Effect of nursery diet protein quality and fish oil supplementation on growth performance, immune response, and gut morphology in starter pigs

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Background: Previous research has demonstrated that nursery diet quality affects growth performance and immune response of starter pigs. This study was conducted to assess the interactive effects of feeding dietary protein quality and oil types on growth performance, immune response and gut morphology in starter pigs.

Methodology: Newly-weaned pigs (21 d age; BW = 6.25±0.83 kg) were randomly assigned to 4 dietary treatments (3 pens/treatment; 10 pigs/pen), as a 2 x 2 factorial design, with differing diet protein quality (**High** vs. **Low**; animal vs. soybean protein), and 5% oil (**Fish** vs. **Corn**). Pigs were fed according to a three phase program; diets fed for 7, 14 and 21 d, respectively. On d 6 and 20 post-weaning, 12 pigs per treatment were vaccinated with 0.5 mg ovalbumin (OVA), 0.5 mg killed *C albicans* (CAA), and 0.5 mg Quil A adjuvant in 1 ml saline. Blood samples were collected at d 20 and 34 for determination of anti-OVA antibodies, and d 22 and 28 for haptoglobin (Hp; acute phase protein; marker for immune response) analysis. The delayed type hypersensitivity (DTH) reaction was evaluated on d 17 and 48, using intradermal injection of OVA and CAA in the ear, and ear thickness was measured at 6h post- injection. Per treatment 6 pigs were euthanized at 2, 4 and 8 wks post-weaning and jejunum samples were collected to evaluate morphology. Data were analysed as a Mixed model procedure of SAS.

Results: There were no significant interactions ($P > 0.10$) between main effects. During the 6 wk starter period, ADG was greater ($P < 0.05$; 422 vs. 385 g) for pigs on **High**; **High** and **Fish** fed pigs had greater feed efficiency (G: F) ($P < 0.05$; 0.750 vs. 0.721, 0.775 vs. 0.695). Diet did not affect ($P > 0.10$) primary or secondary antibody immune response to OVA. At d 17, DTH response to OVA was greater ($P < 0.05$) in pigs fed **Low** diets. At d 48, DTH response to CAA tended to be lower ($P < 0.10$) for pigs fed **Low** and **Fish** diets. The vaccination protocol increased ($P < 0.05$) serum Hp and these were lower in pigs on **Fish** ($P < 0.05$; 0.40 vs. 0.25 g/L). In terms of gut morphology, diet did not impact villus height or villus height: crypt depth ratio; crypt depth was greater (353 vs. 275, 345 vs. 282 μm ; $P < 0.05$) at wk 4 in pigs fed **Low** and **Corn**.

Benefits to the swine industry: These results further strengthen the concept that diet protein quality is critical during the early nursery phase (Phase I & II) to stimulate feed intake and – possibly - the immune system. Thereafter, diet protein quality can be reduced to lower feed costs, while pig performance can be maintained. Independent of diet protein quality, fish oil supplementation improved Gain: Feed, which may be explained by the effect of fish oil supplementation on the pig's response to immunological stress.

Acknowledgements: Swine Innovation Porc, Ontario Ministry of Agricultural, Food and Rural Affairs, and Ontario Pork.

PREVALENCE OF PUTATIVE VIRULENCE GENES IN *HAEMOPHILUS PARASUIS* ISOLATES FROM HEALTHY AND DISEASED SWINE

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BACKGROUND- *Haemophilus parasuis* is a common commensal of upper respiratory tract of pigs of all ages. Under poorly understood conditions it can cause Glasser's disease, a severe septicemic syndrome that is responsible for significant economic loss to swine industry worldwide. There are 15 serovars of *H. parasuis* and some serovars (especially 1, 2, 4, 5, 12, 13, and 14) are thought to have inherently greater virulence potential than others. Many strains are not typable, however, and the virulence potential of some serovars (e.g. sero. 4) is not clear cut.

METHODS- Ninety-four *H. parasuis* isolates (54 from animals with Glasser's disease or pneumonia, 25 from nasal swabs of healthy pigs in farms without Glasser's disease, and 15 reference stains with different reported virulence potential) were evaluated by PCR for the presence of putative virulence genes: *espP2*, *ompP2*, *cdtA*, *cdtB*, *cdtC*, *pilF* and *vapD*.

RESULTS- All 15 reference stains were positive for all of the above genes except serotype 2, 6, 10, and 11, which were *vapD*-negative. As well, 41% of diseased and 60% of healthy pig isolates were negative for the *vapD* gene. The *ompP2*, *cdtA*, *cdtB*, *cdtC*, *pilF* genes were detected in all isolates from the diseased pigs and in most of the isolates from healthy animals. The *espP2* gene was present in all except one diseased pig isolate. Fragment length heterogeneity was detected in the *ompP2* gene with amplicon size ranging between 490 and 510 bp, but there was no correlation between size and the source of the isolates nor was there a correlation between any specific gene and MLVA type.

	<i>vapD</i>	<i>espP2</i>	<i>pilF</i>	<i>cdtA</i>	<i>cdtB</i>	<i>cdtC</i>	<i>ompP2</i>	<i>ompP2</i> 490 bp	<i>ompP2</i> 500 bp	<i>ompP2</i> 510 bp
Healthy N=25	10	25	22	21	23	25	23	13	9	1
Diseased N=54	32	53	54	54	54	54	54	44	6	3

Table 1. Prevalence of putative virulence genes in *H. parasuis* from healthy and diseased pigs

BENEFITS TO THE SWINE INDUSTRY- These studies show that at the present time there is no simple way of identifying *H. parasuis* isolates that have particularly high virulence potential and accordingly, it is probably prudent to consider that all *H. parasuis* may be able to cause disease under the right conditions.

ACKNOWLEDGEMENTS- This work was supported as part of ongoing NSERC and OMAFRA projects.