# DPP2025 Book of Abstracts

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## Theme I: Functionality of the Intestinal Microbiome and Host Response

**1 Searching for the microbes that correlate with pig health, exploring microbial transfer and testing mode of action.** B.P. Willing*, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada*. With increasing pressure on antibiotic use in livestock production, there is a growing interest in finding ways to utilize the microbiome as a means to support healthy pig production. To identify microbial signatures that are associated with health, we, along with many collaborators, have approached this through diverse strategies, including large cohort studies evaluating the microbiome through the lifespan in the context of a natural disease challenge model, vaccine response studies, as well as nation spanning commercial production system surveys. These studies have consistently shown that the pre-weaning microbiome is a very good predictor of subsequent immune function and growth. We have also identified the wild microbiome as a potential source of beneficial microbes for the domestic pig through fecal transplant studies. We have found that wild boar derived microbes are more successful in colonizing the weaned piglet than sow derived microbes, and result in reduced levels of pro-inflammatory cytokines. These studies suggest that there may be an opportunity to introduce cultured wild boar derived microbes to commercial pigs to enhance health. Using gnotobiotic piglet and mice models, we are starting to unravel how these early life microbial communities, and their interactions with diet and other stressors can impact on the development of the immune system to support subsequent disease resilience. We employ metabolomics and transcriptomics to gain insight into modes of action of dietary and microbial interventions in early life and the weaning transition. The take-home message of this presentation is that there is still significant opportunity in harnessing the microbiome for enhanced pig production, but significant opportunities for continued discovery lay ahead. **Key Words:** diet-microbe-host interactions microbial metabolites microbial programming

**2 EU Circles project: Machine Learning Approaches to Multi-Kingdom Gut Microbiota Reveal Key Predictors of Piglet Growth During the Nursery Phase.** F. Correa*[1], D. Luise[1], G. Palladino[2], F. Palumbo[1], D. Scicchitano[2], G. Babbi[2], A. Castagnetti[3], M. Soverini[3], S. Rampelli[2], M. Candela[2], P.L. Martelli[2], and P. Trevisi[1], [1]*Department of Agricultural and Food Sciences, University of Bologna, 40127 Bologna, Italy*, [2]*Department of Pharmacy and Biotechnology, University of Bologna, 40126 Bologna, Italy*, [3]*Wellmicro srl, 40128 Bologna, Italy*. During the nursery phase the gut microbiota plays a central role in shaping growth performance by regulating nutrient metabolism, immune responses, and resilience to environmental stressors. While most studies have focused on bacterial microbiota, the roles of other microbial kingdoms, such as viruses, fungi, and other microorganisms, remain underexplored. This study employed a multi-kingdom metagenomic approach to investigate the associations between fecal microbiota composition and growth during the nursery phase. A cohort of 304 pigs from the same farrowing unit were monitored. Pigs were weighed at weaning (T0) and divided into two weaning units, classified as low- and high-performance groups based on their historical growth metrics. Weights were recorded after seven days post-weaning (T1) and during the final week before moving the pigs to the growing unit (T2). Fecal samples were collected at each timepoint for metagenomic profiling. An R-based machine learning pipeline that employs the Random Forest algorithm (VPR1.0) to analyze microbial and environmental features was used to predict the weight gain. The obtained model was cross-validated on an external independent dataset, achieving a Pearson correlation of 0.542 ($P < 0.001$) with the validation dataset, highlighting the predictive potential of microbial and contextual factors. Key factor associated with growth included *Lactiplantibacillus plantarum* (r = -0.25, $P < 0.001$), *Lactobacillus* phage phiAQ113 (r = -0.22, $P < 0.001$), *Streptococcus agalactiae* (r = 0.22, $P < 0.001$), and *Desulfovibrio piger* (r = 0.15, $P < 0.001$), alongside farm origin, which showed a strong positive correlation with the predicted outcome (r = 0.34, $P < 0.001$). This study underscores the value of a multi-kingdom perspective in understanding the gut microbiota's role during weaning, offering a holistic view of microbial and environmental factors influencing piglet growth. **Key Words:** metagenomic, random forest

**3 Gut Microbiota-host cells interaction in Health and Diseases.** H. M. Blottière*[1,2], [1]*Nantes Université, INRAE, UMR 1280 PhAN, F-44000, Nantes, France*, [2]*Université Paris-Saclay, INRAE, MetaGenoPolis, MGP, F-78350, Jouy-en-Josas,*

*France*. The intestinal microbiota is considered as a true organ, playing major roles for its host. Dysbiosis has been shown in a number of diseases including inflammatory bowel diseases, metabolic disorders but also neurological disorders. However, little is known about the underlying mechanisms. Few specific effectors interacting with the mammalian host have been identified. Bacterial metabolites, notably short chain fatty acids (SCFAs), branched-chain amino acids, tryptophan metabolites including indoles and bile salts have been shown to be key mediators between the bacteria and the host. In the case of SCFAs, they provide energy to colonic epithelial cells, control their proliferation, exert anti-inflammatory effects and control motility. In fact, they act as key regulators of cell signaling, acting on specific receptors such as G-protein coupled and nuclear receptors, but also as regulator of the acetylation of certain proteins, inhibiting the histone deacetylase that regulates gene expression. Among the factors that limit our current knowledge of the mechanisms of interaction between the gut microbiota and host cells, the difficulties in culturing most commensal bacteria and the complexity of the culture medium are of particular importance. The development of metagenomic approaches has been a decisive step towards understanding the functions of the intestinal microbiota. We have therefore developed a high-throughput functional metagenomic approach to overcome these limitations and identify bacterial genes and compounds regulating key signaling pathways or effector genes in mammalian immune, intestinal or liver cells. Genomic and metagenomic libraries were constructed by cloning large DNA fragments into a fosmid vector and using *E. coli* as the host. A high-throughput screening platform was established, enabling the identification of clones modulating pathways and genes in mammalian cells. Although the host was a gram-negative bacterium, it was possible to identify genes derived from the three main phyla present in the gut, namely *Bacillota*, *Bacteroidota* and *Actinomycetota*. This functional metagenomic strategy allowed to characterize new genes from commensal bacteria and effector molecules that have an impact on the host's cellular functions, with a potential influence on mammalian health. **Key Words:** Metagenomics host cell functions HTS

**4 Litter Origin is associated with Gut Microbiome Composition During Tail-Biting Outbreaks in Growing-Finishing Pigs.** Sudario Roberto Silva Junior*[1], Courtney Archer[1], Lee Johnston[1,2], Yuzhi Li[1,2], and Andres Gomez[1], [1]*Department of Animal Science, University of Minnesota, St. Paul, MN, USA*, [2]*West Central Research and Outreach Center, University of Minnesota, Morris, MN, USA.* This study aimed to investigate the impact of litter origin on the gut microbiome during tail biting outbreaks (TBO) in undocked pigs. Pigs (N=126; 10 weeks old) were assigned to one of three treatments: Littermates (LM: same sow), Non-Littermates (N: each pig from a different sow), and Half-Littermates (H: 4 from one sow, 3 from another sow) with 7 pigs/pen. Tail injuries were scored weekly (0–4), and a TBO was defined as ≥1 pig in a pen having a score ≥2. During a TBO, fecal samples were collected from all pigs in affected pens. Deoxyribonucleic acid was extracted from fecal samples and the V4 region of the 16S rRNA bacterial gene was amplified and sequenced on the MiSeq platform. Taxonomic abundances were determined and analyzed using R packages, and functional profiles predicted with PICRUSt2. Half-littermate pigs, which had higher tail injury scores, showed lower Shannon diversity and richness than N pigs (Kruskal-Wallis, $P = 0.015$); but were similar to LM pigs. Beta diversity based on Bray–Curtis distances indicated differences in microbiome composition based on litter origin (PERMANOVA, $P < 0.01$, $R^2 = 0.04$), which was confirmed via partial least squares discriminant analysis (AUC P > 0.91). Lower abundances ($P < 0.05$) of C*lostridium butyricum*, *C. moniliforme* and *C. celatum*, which are typically involved in short-chain fatty acid production, were observed in H and LM pigs. These pigs also showed lower abundance of *Romboutsia ilealis* and *Turicibacter*, which are involved in amino acid fermentation and bile acid metabolism. Functional predictions indicated that H and LM pigs indeed harbored lower abundance of genes predicted to be involved in butanoate and propanoate metabolism, histidine synthesis, and metabolism of purines and pyrimidines. These findings suggest that litter origin is associated with gut microbiome composition and potential function during TBO. **Key Words:** litter origin microbiome tail biting

**5 Exploring the Gut Microbiota's Impact on Sow Performance: Links Between Performance, Stage in Reproductive Cycle, and Key Factors in a European Sow Study.** M. Weiss*[1,2], G. A. Vestergaard[2], S. E bohdidi[2], L. H. B. Hansen[2], T. T. M. Knudsen[2], and D.S. Nielsen[1], [1]*University of Copenhagen, Department of food Science, University of Copenhagen, 1958 Frederiksberg, Denmark*, [2]*Novonesis, Novonesis, Animal Biosolutions, Biologiens vej 2, 2800 Kongens Lyngby, Denmark.* The gut microbiota significantly impacts sow health and performance, as shown by comparing individual sow performance on similar farms. However, factors like geography, diet, and reproductive stage may limit the predictive power of machine learning. The objectives of this study were to determine if a sow microbiota can be defined and show associations between the microbiota and various sow characteristics, including reproductive stage, parity, use of probiotics, and performance. This would indicate that it is possible to predict performance based on microbiota data from conventional farms. Fecal samples were collected from over 200 multiparous sows across major swine producing countries in Europe on day 109 of gestation and day 21 of lactation (min. 12 sows per farm). Six out of 16 farms administered probiotics. Samples were analyzed using 16S rRNA gene long-read amplicon sequencing. The reproductive stage was more associated with the sow gut microbiota than country, as shown by microbiota composition by comparing robust Aitchison distances with PERMANOVA testing (reproductive stage: $P = 0.001$, F = 33.14; country: $P = 0.001$, F = 11.95). Probiotics ($P = 0.001$, F = 4.62) and performance (weaned pigs/sow/year, $P = 0.001$, F = 4.16) also modulated the microbiota, while parity had a smaller, yet still significant impact ($P = 0.002$, F=1.87). During the reproductive stage, a core microbiota was identified (prevalence above 90%, abundance > 0.5% of total reads), comprising 39 species on day 109 of gestation and 24 species on day 21 of lactation, with an overlap of 20 shared species. Notably, most species unique to each stage were differentially abundant between the two sampling periods (17/23 with $P < 0.05$). In conclusion, the reproductive stage of European sows has a greater impact on gut microbiota composition than country or parity, with distinct core microbiota at each stage. Furthermore, microbiota composition is strongly correlated with probiotics and performance. **Key Words:** sow gut microbiota, performance, reproductive cycle

**6 Advances toward commercial use of fecal microbiota transplantation to mitigate weaning stress in pigs.** Paul Oladele, Wenxuan Dong, Brian Richert, and Timothy Johnson*, *Purdue University, West Lafayette, IN, USA.* Weaning is a critical and stressful period for piglets, commonly resulting in post-weaning diarrhea (PWD). In some studies, fecal microbiota transplantation (FMT) reduced diarrhea and improved piglet growth. To enhance FMT feasibility for use on commercial farms, we conducted studies to improve donor selection, fecal bacterial community preservation, and large-scale administration. To identify optimal FMT donors, fifteen fecal samples from weaners, growers, and sows from the same farm were selected for an anaerobic in vitro fermentation study, in a 2x2 design, with and without enterotoxigenic E. coli (ETEC) F18 and in ileal and colonic simulating media. Samples from growers inhibited ETEC F18 ($P < 0.05$) and produced higher levels of acetate and butyrate ($P < 0.05$). Next, we investigated the impact of lyoprotectants on bacterial viability and change in community structure after lyophilization. Trehalose and maltodextrin best preserved both bacterial viability and community composition in fecal samples. Two in vivo experiments were conducted to compare the colonization efficiency of three FMT administration methods—oral gavage, rectal gavage, and lyophilized in-feed. In the first experiment, all FMT groups (n=10 individually housed piglets/group) had significantly higher body weight gain ($P < 0.05$) on day 3 compared to no FMT. On day 4, colon crypt depth was increased in all FMT groups, but an increase in ileal villus length was only observed in the in-feed group. The in-feed group had higher fecal microbiome alpha diversity ($P < 0.05$) on day 5. In the second experiment (8 pens of 6 weaned and transported piglets per group), FMT reduced the diarrhea index ($P < 0.05$) on day 5, but did not increase weight gain ($P > 0.05$). In conclusion, in-feed FMT shows promise to alleviate weaning stress and to improve growth. Screening donor fecal samples for specific microbiome traits, such as pathogen inhibition and fermentative capacity, could enhance and standardize FMT outcomes. **Key Words:** microbiota colonization, post-weaning diarrhea, bacterial viability

**7 Carbohydrate and nitrogen requirements for optimizing hindgut microbiome in pigs.** Ehsan Khafipour[1], Sandra Paredes[1], Qiong Hu*[1], Maria Sardi[2], and Ali Naqvi[2], [1]*Cargill Animal Nutrition and Health, Minneapolis, MN*, [2]*Cargill, Minneapolis, MN*. The composition of carbohydrates (CHOs) and nitrogen (N) sources is critical for maximizing fermentation and production of microbial crude protein (MCP) and volatile fatty acids (VFA). However, pigs have a low capacity to benefit from MCP while they can absorb VFA, which contributes up to 30% of their energy requirement. Previous research indicates that the microbiome needs to be kept in a N-deficient state compared to CHOs availability to promote VFA production while limiting MCP synthesis. We performed five nursery studies with varying levels of structural CHO (CHO STR) and hindgut fermentable protein (GUT FP) to assess the composition, functionality, and robustness of pig fecal microbiome. The CHO STR content of the diets varied from 3-6%. GUT FP of diets was the sum of all ingredients' GUT FP and ranged from 1-1.2%. Fecal samples for microbiome analyses (n= 1 pig/pen, 8-12 pens/treatment depending on the study) were collected on d7 and 14 post-weaning and subjected to shotgun metagenomics. Microbiome data were center-log-ratio transformed and analyzed for diversity and composition, and microbial community interaction networks. Increasing CHO STR from 3% to 4.5% and above reduced (3.88 vs. 4.18; 4.20 vs. 4.37, 4.30 vs. 4.64, $P < 0.05$) fecal microbiome diversity in three studies, tended to reduce in one study (4.59 vs. 4.75; $P = 0.06$) but had no impact in the last study. This reduction in diversity was accompanied by the promotion of a leaner (reduced edge density) but more connected (increased positive edge percentage) microbial networks. Increase in GUT FP levels did not affect the strength of the microbial network. Numerically higher abundance and centrality of a subset of influential species with no clear pattern was observed with higher CHO STR. No clear pattern of bacterial species associated with GUT FP was found. This suggests that maintaining a robust hindgut microbiome can be achieved by promoting fibrolytic activity via increasing dietary CHO STR level. **Key Words:** Hindgut microbiome, Gut fermentable protein, Structural carbohydrates

**8 Fecal filtrate transplantation and dietary fibre supplementation as alternatives to veterinary antimicrobials.** A. Middelkoop*[1], J. Priem[1], C. Larsen[2], T. Thymann[2], and F. Molist[1], [1]*Schothorst Feed Research, Meerkoetenweg 26, 8218 NA Lelystad, The Netherlands*, [2]*University of Copenhagen, Dyrlægevej 68, 1870, Frederiksberg C, Denmark*. Early-life modulation of the piglet microbiome has been shown to affect post-weaning diarrhea. We aimed to investigate the effects of fecal filtrate transplantation (FFT) in newborn piglets (n=32 litters/treatment) and the effects of an alfalfa-enriched diet in the first two weeks post-weaning (n=32 pens/treatment; 6 piglets/pen in 1:1 sex ratio) on weaner pig health and performance in a 2 x 2 factorial design. Each piglet in the litter was inoculated from day 1 to 6 of life with 6 ml/day of either fecal filtrate (FFT+) or SM-buffer (FFT-). FFT+ piglets received an oral dose of fecal filtrate, prepared by dilution and micro-filtration of pooled pathogen-free faeces derived from 15 lactating sows on-farm. In addition, piglets were fed either a low-fibre (Fibre-) or a high-fibre (Fibre+) phase 1 weaner diet (day 0-14 post-weaning), of which the latter contained 5% alfalfa. All piglets were fed the same creep feed (day 7 until weaning) and phase 2 weaner diet (day 14-35 post-weaning). Fecal consistency (score 2-9) was scored daily in phase 1 and 3x a week in phase 2 on pen level. Data were analyzed with ANOVA. Individual antimicrobial usages were recorded and analyzed with Chi-square. Post-weaning piglet morbidity was 6.8% and mortality was 1%. No interactions among FFT and dietary fibre supplementation were observed. FFT+ reduced antimicrobial usage versus FFT- by 7% ($P < 0.01$) and tended to improve fecal consistency in week 5 post-weaning (6.0 vs. 5.8±0.09; $P < 0.10$). Fibre+ improved the fecal score in the overall 5-week post-weaning period versus Fibre- (5.5 vs. 5.3±0.07; $P < 0.10$). A lower percentage of piglets required antimicrobial treatment in Fibre+ (3.2%) versus Fibre- (10.4%; $P < 0.01$). No effects of the two factors were seen on body weight and feed efficiency. To conclude, pre-weaning fecal filtrate transplantation and post-weaning dietary alfalfa supplementation may have the potential to reduce antimicrobial usage in weaned piglets via improved gut functionality. **Key Words:** Fecal filtrate transplantation, dietary fibre, post-weaning diarrhea

## Theme I Posters: Functionality of the Intestinal Microbiome and Host Response

**9 Exploring the impact of alternative carbohydrate sources on the gut microbiota of pigs.** D Schokker*[1], F Veldkamp[2], S van Hemert[1], N Stockhofe[1], JMJ Rebel[1,3], and IC de Jong[2], [1]*Wageningen Bioveterinary Research, Lelystad, Flevoland, The Netherlands*, [2]*Wageningen Livestock Research, Wageningen, Gelderland, The Netherlands*, [3]*Wageningen University, Wageningen, Gelderland, The Netherlands*. The gut microbiota is crucial for pig health and welfare. This study examines the impact of different sugar sources on the gut microbiota. Three treatments were tested (all isoenergetic diets), a control diet (CON), a former food products diet (FFP, simple carbohydrates), and a sugar beet pulp diet (SBP, complex carbohydrates). Sixty pigs were sampled at days 7 and 42 post-weaning, with 10 jejunum and colon samples collected per treatment group at each time point. Microbiota profiles were determined by long-read sequencing, use our in-house pipeline for taxonomy, and subsequently utilized the Cazy database for gene families involved in carbohydrate metabolism. The jejunal microbiota data showed significant day ($P = 0.001$) and treatment ($P = 0.021$) effects. At day 7, *Turicibacter* abundance was lower in SBP (0.02%) compared to CON (0.05%) and FFP (0.05%). The colon microbiota data showed a significant interaction effect between day and treatment ($P = 0.007$). At day 7 in colon, *Lachnospira* was higher in SBP (0.03%) compared to CON (0.003%) and FFP (0.001%), whereas at day *Lachnospira* was 42 SBP 0.04% compared to CON (0.01%) and FFP (0.01%). MGX results in colon corroborated that *Lachnospira eligens* was significantly higher ($P < 0.001$) in SBP (12.0%) compared to CON (3.3%). At day 42, *Lactobacillus delbrueckii* was significantly higher ($P < 0.001$) in SBP (0.37%) compared to CON (0.07%). Significant enrichment ($P < 0.001$) of Polysaccharide Lyase Family 9 (PL9) was observed, PL9 acts on pectins. On day 7, CON had a relative abundance of 0.09% and SBP 0.25%. At day 42, CON remained at 0.09%, while SBP increased to 0.31%. Integrating taxonomic and functional data showed the link between *L. eligens* and PL9. These sources modulate microbial diversity and enzyme activity, particularly enhancing beneficial genera like *Lachnospira* and *Lactobacillus*. This study demonstrates that sustainable sugar sources positively influence gut microbiota in pigs. **Key Words:** gut microbiota, carbohydrate source, metagenomics

**10 Impact of dietary fibers on bacterial community composition and volatile fatty acids production in pigs.** O. Munezero*[1], E. M. Due[2], N. K. Gabler[2], T. E. Burkey[1], and S. C. Fernando[1], [1]*University of Nebraska-Lincoln, Lincoln, Nebraska, USA*, [2]*Iowa State University, Ames, Iowa, USA*. Dietary fibers provide an energy source for gut microbial fermentation, and in turn, provide volatile fatty acids (VFA) and other metabolites for host metabolism and function. However, the effects of dietary fiber on bacterial community composition and VFA production are poorly understood as these factors have not been investigated simultaneously. Here we investigated the impact of dietary fibers on bacterial community composition and VFA production. The 450 weaned pigs, weighing 5.6 ± 0.66 kg each, were randomly assigned to one of five dietary treatments (Control diet, 6% wheat bran, 6% beet pulp, 8.29% corn DDGS, 0.20% Tributyrin) in a 42-day feeding trial. Each treatment had 9 pens with 10 pigs per pen. Fecal samples for microbiome and VFA analysis were collected on day 14 and 42. The bacterial community of the fecal samples was assessed by sequencing the V4 region of the 16S rRNA gene. There was no effect of dietary fiber treatment on the Shannon diversity and observed amplicon sequence variants (ASVs), except for Beet Pulp vs tributyrin which showed a significant difference ($P = 0.048$). However, β-diversity analysis identified distinctive clustering among treatments with both unweighted UniFrac and Bray Curtis dissimilarity matrix showing significant differences in bacterial community composition among treatment groups and between time points ($p < 0.05$). Differential analysis between different fiber sources identified several ASVs that were significantly different between treatments and included ASVs belonging to *Lactobacillus*, Clostridia UCG-014, *Limosilactobacillus*, and *Ruminococcus*. Significant associations were also observed between bacterial taxa and VFAs. *Roseburia* was associated with increased propionic acid. Additionally, *Syntrophococcus* was associated with both propionic and valeric acids. In conclusion,

supplementation of swine diets with different dietary fibers affected the bacterial community composition, which in turn led to different VFA profiles. **Key Words:** dietary fibers, microbiome, volatile fatty acids

**11 A balancing act: the crucial role of the microbiome in antimicrobial-free fed piglets.** C. Turni* and V.H. Tran, *Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, St Lucia, Australia*. Early weaning is a stressful event occurring under commercial practices associated with social and environmental challenges and an abrupt change from milk to a solid feed. There is a period of adaptation associated to several hours of fasting, compromising gut health and the microbiome-host balance. Early in life, gut microbiota is influenced vaginal and skin microbiota and colostrum (farrowing), and naturally occurring faecal transfers sow-piglet (lactation). The dominance of *Bacteroidaceae*, *Clostridiaceae*, *Lachnospiraceae*, *Lactobacillaceae*, and *Enterobacteriaceae* is associated with the use of mono-, di- and oligo-saccharides. In the colon and caecum Bacteroidetes (38.7%) and Firmicutes (54%) are the dominant bacteria during lactation. Post-weaning there are significant changes with increasing numbers of *Prevotellaceae* and *Ruminococcaceae*. In the ileum *Lactobacillus*, *Streptococcus* and *Clostridium* become more abundant related to diet. There is a gradual shift up to 22 weeks of age with Firmicutes increasing, while Bacteriodetes degrease. Fermentable fibre in post-weaning feeds stimulate gut fermentation and promote proportion of fibre-degrader *Bacteroidetes* up to 59.6%, while oligosaccharide utilisers *Bacteroides*, decrease. High crude protein levels in post-weaning feeds can contribute to diarrhoea due to undigested proteins fostering pathogenic bacteria in the gut such as the *Enterobacteriaceae* family. Additional supplements to early feeds, such as fermented or extruded wheat bran, probiotics and prebiotics, and amino acids, showed positive improvements in gut microbial profile and morphology. Diets high in pectin and fish protein may reduce *Lactobacillus*, increasing susceptibility to pathogens, while plant-based proteins helped to lower *E. coli* counts, albeit with some impact on growth. In summary, formulating post-weaning feeds should be carefully examined to provide adequate nutrition but also maintain a healthy gut microbial profile. **Key Words:** weaning, gut health, microbiota

**12 Feed intake modulates fecal microbial communities in weaning pigs.** H Tran*, AJ Mercado, B Rimal, and B de Rodas, *Purina Animal Nutrition, Gray Summit, MO, USA*. The objective of this trial was to evaluate the impact of intake behavior on growth performance and the fecal microbiome of nursery pigs and identify key bacteria markers associated with better feed consumption. Thirty pigs (PIC; BW = 6.35 kg; 20 d old) were used. From d 0-10, pigs were individually penned and fed a common starter diet, with daily feed intake recorded. Individual pig BW was recorded on d 0, 4, and 10. ADFI during d 0-4 (0.13±0.05 kg) was used to retroactively categorize pigs into eating groups, including non-eaters (ADFI < 0.08 kg), average (AVE; 0.08 ≤ ADFI ≤ 0.19 kg), and eaters (ADFI > 0.19 kg). On d 10, pigs were moved to group pens by treatment and fed a common diet until d 28. Fecal samples were collected on d 0, 4, 10, and 28 for 16S rRNA sequencing (V3-4, Illumina MiSeq). From d 0-4 and d 0-10, eaters had greater ADFI, ADG, and BW compared to non-eaters ($P < 0.01$). Regarding microbial diversity, beta diversity shifted significantly over time ($P < 0.01$). On d 4, non-eaters had fewer observed microbial features versus the AVE ($P < 0.001$), but no significant difference from the eaters ($P = 0.14$). LEfSe analysis identified specific bacterial genera associated with eating behavior. Particularly, *Prevotellaceae NK3B31*, *Limosillactobacillus*, *Faecalibacterium*, and *Lactobacillus* were enriched in the eaters (LDA > 4, $P < 0.01$), while *Lachnoclostridium* and *Escherichia-Shigella* were associated in non-eaters (LDA > 4; $P < 0.01$). Additionally, eaters had lower total propionate producers (*Bacteroidaceae, Ruminococcaceae, Acidaminococcaceae,* and *Christensenellaceae; P* < 0.05), but greater total carbohydrate degraders and lactate producers (*Lactobacillaceae, Ruminococcaceae*, and *Prevotellaceae; P* < 0.01) versus non-eaters on d 4. The results suggest that early feed intake differences influence nursery performance and gut microbiome composition. Higher feed intake post-weaning may accelerate microbiome maturation, while lower feed intake is associated with increased opportunistic bacteria. **Key Words:** feed intake, fecal microbiome,

weaning pig

**13 Unlocking the power of novel precision biotic on microbiome-gut-brain axis modulation: Enhancing undocked tail pigs' resilience to social stress.** W. Ren*[1], C. Gruber[2], I. Gradner[2], J. Howard[2], N. Reisinger[2], R. Argamasilla[3], and E. Perez-Calvo[3], [1]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Bazhou, China*, [2]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Tulln, Austria*, [3]*dsm-firmenich, Animal Nutrition and Health, Kaiseraugst, Switzerland*. Microbiota-gut-brain axis and its influence on pigs' behavior has been recently explored suggesting that there may be a link between specific microbiota functions and the development of manipulative behaviors. This study aimed to evaluate the effect of a novel precision biotic in post-weaning piglets under social stress, by assessing lesion severity, stress hormone levels and blood metabolite pathways. Seventy-two weaned pigs with undocked tails and equal numbers of male and female were allocated to two dietary treatments: either a control or control + 250 ppm PB diet (PB) for 21 days, after 7 days adaptation post-weaning. The social stress challenge was conducted at d 19 by regrouping pigs within the same treatment. Lesion scores on the body, tail, and ear were evaluated. Cortisol (COR) and adrenocorticotropic hormone (ACTH) levels were measured at saliva and plasma targeted metabolome determination. Growth performance was also monitored. The results were analyzed by JMP and the significance was defined at $P < 0.05$. Severe body and tail lesions increased from 0% to 73%, and 0% to 22%, respectively, after social stress challenge. Piglets receiving PB showed a significant reduction in body lesion scores (-17%, $P = 0.03$) and a numerically reduced tail lesion score after the social stress. Numerically lower COR and significantly lower ACTH levels (22.99 *vs.* 33.49 ng/mL, $P = 0.04$) were observed in PB animals, indicating enhanced stress resilience. Plasma analysis showed enriched gamma-aminobutyric acid (GABA) metabolic pathways in PB treatment, indicating increased microbiota GABA production and transportation. PB supplementation improved feed conversion ratio (1.37 *vs.* 1.57, $P = 0.03$) and average daily gain (400 *vs.* 362 g/d, $P = 0.05$). In conclusion, PB supplementation promoted pig resilience to social stress by modulating the GABA production potentially generated by the gut microbiota, which consequently had a positive effect on growth performance. **Key Words:** Precision biotic, Microbiota-gut-brain axis, Animal welfare

**14 Using a multi-strain *Bacillus* spp. probiotic to improve growth performance and modulate the gut microbiome in weaned pigs.** E. Vinyeta*[1], D. E. Velayudhan[1], Q. Wang[2], S. Bialkowski[2], J. Walker[2], C. Shen[3], K. Rassmidatta[4], and Y. Ruangpanit[4], [1]*Danisco Animal Nutrition & Health (IFF), Oegstgeest, The Netherlands*, [2]*Health & Biosciences (IFF), Wilmington, Delaware, USA*, [3]*IFF Nutrition & Biosciences, Brabrand, Denmark*, [4]*Kasetsart University, Kamphaengsaen Campus, Nakhon Pathom, Thailand*. This study evaluated the effect of a *Bacillus* spp. multi-strain probiotic fed to pigs from day 1 to 42 post-weaning on growth performance and fecal microbiome. A total of 400 weaning piglets (initial pig body weight of 7.2 ±1.5 kg) were assigned to 2 dietary treatments, 20 pen replicates/treatment, and 10 piglets/ pen (1:1 male: female). Treatments comprised of 1) rice-corn-soybean meal-whey-based control diet; and 2) control diet supplemented with 3-strain *Bacillus amyloliquefaciens* providing 3.0 × 10^8 cfu/kg of feed. Diets were fed as mash in 2 phases (1-14 and 14-42 days). Pigs were weighed individually, feed intake was recorded per pen at the end of each phase, and average daily gain (ADG), average daily feed intake and feed conversion ratio (FCR) were calculated. Fresh fecal samples were collected (2 pigs/pen, 10 pens/treatment) on days 0, 21 and 42 to determine the microbiome population using 16S amplicon sequencing. Performance data were analyzed using Fit Model (JMP 16.1) with pen as experimental unit. Differences in microbial community compositions between treatments were assessed using PERMANOVA with sex as a random effect. Differentially abundant microbial taxa were identified using DESeq2 controlling for sex differences. The ADG increased ($P < 0.05$) during days 1-14 and 1-42 with probiotic supplementation. The FCR was lower ($P < 0.05$) during days 1-14 when fed probiotics. Overall mortality was reduced ($P < 0.05$) with probiotic supplementation (1.6 vs 1.0%). Microbiome analysis showed that the microbial communities were similar between treatments on day 1, then gradually shifted over time ($P < 0.05$). Several genera

including *Segatella* on day 21, *Clostridium, Faecalibacterium and Xylanibacter* on day 42, were significantly more abundant ($P < 0.05$) in the probiotic-fed diet. In conclusion, multi-strain probiotics improved weight gain and feed efficiency of weanling pigs, reduced mortality and improved the abundance of certain beneficial microbial taxa. **Key Words:** weaned pig, *Bacillus* spp probiotic, performance and gut microbiome

**15 A prairie province wide survey of Canadian swine fecal microbiomes reveals a high degree of variation between barns.** M. B. Rogers*[1], B. McCuaig[2], E.L. McCarthy[2], S. L. Saundh[2], R. de Almeida Mesquita[2], T. Prisnee[3], M.O. Wellington[2], A. K. Agyekum[2], J. Harding[4], B. Willing[3], M. Links[2], and A. Van Kessel[1]. [1]*Vaccine and Infectious Disease Organization, University of Saskatchewan, Saskatoon, Saskatchewan, Canada,* [2]*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada,* [3]*Faculty of Agricultural, Life and Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada,* [4]*Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, Saskatchewan, Canada*. With the objective of linking health and productivity outcomes in the Canadian pork industry with swine gut microbiome signatures we have embarked on a multi-province and multi-farm longitudinal study of sows and piglets from early lactation through near market. Fecal samples were collected from 603 pigs (298 females, 305 males) across 15 cohorts (farms) over 225 days, including three collections pre-weaning and three post-weaning. Of the 15 farms, 10 used conventional and 5 used raised without antibiotics management strategies. Fecal swabs were also collected from 156 maternal sows (mean 10.4 per barn). 16S rRNA gene amplicons (V3-V4) were generated from fecal samples and fecal swabs, amplicon sequence variants (ASVs) were predicted using DADA2 in R, and taxonomy was assigned with RDP. Diversity metrics and taxonomic profiles were generated using vegan and phyloseq. Longitudinal variation (variation between collection points) in piglets had the greatest effect on microbiota variation (ADONIS2; $R^2$=0.198; $P < 0.001$), followed by farm ($R^2$=0.09; $P < 0.001$). Variables like diet, health scores, sex, and production rank showed no significant effect while accounting for the above variables and interactions. To assess sow-to-pig transmission, we contrasted cao beta-diversity distances between sow-piglet pairs and random piglets in the same cohort. In most cases (3/156 exceptions), no significant differences were found ($P < 0.05$), with nearly identical mean cao distances (2.908 vs. 2.890) between sow piglet pairs and random piglets respectively. In conclusion, similar to the human microbiome, geography and temporal succession are primary among variables shaping the pig microbiome. Although we cannot reject the hypothesis that the maternal fecal microbiome shapes the microbiome of the piglet, we find no evidence for it in this study. The level of variation observed suggests that pig management aimed at microbiome optimization may need to be customized to the farm. **Key Words:** pig gut microbiome, longitudinal variation, variation across farms

**16 Exploring the interplay between mucin O-glycans and the gut microbiome in pigs.** L Richardson*, L Kautto, and N Packer, *ARC ITTC FAAB, School of Natural Sciences, Faculty of Science and Engineering, Macquarie University, North Ryde, Sydney, NSW 2109, Australia*. The digestive tract is coated with mucus, a lubricant that protects epithelial cells from mechanical forces such as digestion and serves as a barrier against pathogens by trapping microorganisms. Mucins are glycoproteins that form a critical component of mucus, and they are decorated with complex carbohydrates known as mucin O-glycans. Mucin O-glycans serve as both binding sites and nutrient sources for gut bacteria, directly influencing the composition of bacterial communities in the digestive tract. For instance, *Akkermansia muciniphila* binds to O-glycan structures to aid its attachment to the mucus layer and promote mucus turnover. In contrast, *Bacteroides fragilis* metabolises mucin-derived sugars, enabling cross-feeding interactions with other microbes, such as *Faecalibacterium prausnitzii*. This research investigated the relationship between mucin O-glycans and the gut microbiome in pigs. By analysing the mucus and luminal microbiomes along with the structure of mucin O-glycans across the jejunum, ileum, and colon, we aimed to uncover how regional variations in O-glycan structures correlated with distinct microbial communities in pigs. Our results showed

increased sulphation as you go down the colon, which positively correlated with an increase in *Lactobacillus* as well as other species of bacteria. These findings have significant future implications for dietary strategies involving probiotics, prebiotics, and fecal microbiota transplants, offering a foundation for approaches to improving gut health in pigs. **Key Words:** mucin, O-glycans, microbiome

**17 Effect of *Saccharomyces cerevisiae boulardii* CNCM I-1079 on bile acid signaling and fecal microbiota composition in post-weaning piglets.** C. Achard[1], F. Bravo de Laguna[1], A. Gavaldá-Navarro[2], I. Alvarez-Acero[3], F. Villarroya[2], S. de Pascual-Teresa[3], E. Chevaux[1], D. Saornil[1], M. Castex[1], and I. R. Ipharraguerre*[4], [1]*Lallemand SAS, Blagnac, France*, [2]*Department of Biochemistry and Molecular Biomedicine, Faculty of Biology, Barcelona, Spain*, [3]*Institute of Science and Technology of Food and Nutrition (ICTAN-CSIC), Madrid, Spain*, [4]*Institute of Human Nutrition and Food Science, University of Kiel, Kiel, Germany*. Beneficial effect of the live yeast *Saccharomyces cerevisiae boulardii* CNCM I-1079 (SB) administered to weanling piglets was previously shown on performances: better growth at day 14 post-weaning was associated with altered bile acids (BA) metabolism (Bravo de Laguna et al, DPP 2022). The objective of this follow-up study was to investigate whether BA profile modulation was linked to changes in fecal microbiota. 28-day old weaned piglets were fed either Control or SB supplemented (2 x $10^9$ CFU/kg until day 15 post-weaning, $10^9$ CFU/kg until day 36) diets. Fecal samples were collected at days 6 and 36 (D6, n = 58; D36, n = 39) for microbiota analyses via 16S rRNA gene sequencing. Fecal BA profiles were analyzed at the same days. Bioinformatics and statistical analyzes were performed with R packages (DADA2, Vegan, lmertest, MixOmics). The fecal microbiota was shaped by the age of the piglets. Bacterial density increased with age ($2.60\times10^{11}$ vs $6.72\times10^{11}$ 16S gene copies/g of feces, $P < 0.001$). At D6, microbial richness was more homogeneous in SB group (Levene test, $P < 0.05$). Alpha-diversity diminished with age, in SB group only (Kruskal-Wallis test, $P < 0.01$). SB effect on fecal microbiota was evidenced using Partial Least Square Discriminant Analysis. Estimated model performances allowed the selection of 39 and 49 discriminant ASVs (Amplicon Sequence Variants), respectively at D6 and D36 (perf = 0.36 and 0.27, 100 cross-iterations). Regularized Canonical Correlation analyses positively linked fecal secondary BA to *Prevotellaceae*, *Succinivibrio*, *Acidaminococcus* and *Selenomonadaceae* ASVs, while negative correlations were shown with *Oscillibacter*, *Campylobacter*, *Treponema*, *Fibrobacter* and *Coprococcus* ASVs (cutoff=0.5). One selected discriminant ASV (unknown *Lachnospiraceae*) was also associated with low Chenodeoxycholic/Hyocholic acids ratio. Our results suggest that SB may modulate microbial conversion and intestinal uptake of primary BA, contributing to better post-weaning growth. **Key Words:** live yeast, probiotics, bile acids metabolism

**18 Blood metabolic and fecal microbial profile of sows fed high and low fiber diets with hessian or straw enrichment prior to farrowing.** E. A. Soumeh*[1], S. E. James[2], R. J. Moore[3], L. M. Staveley[4], K. J. Plush[4], and T. L. Nowland[2], [1]*School of Agriculture and Food Sustainability, The University of Queensland, Gatton Campus, Gatton, QLD, Australia*, [2]*Aquatic and Livestock Sciences, South Australian Research and Development Institute, Roseworthy, SA, Australia*, [3]*School of Science, RMIT University, Bundoora West Campus, Bundoora, VIC, Australia*, [4]*SunPork Group, Eagle Farm, QLD, Australia*. Dietary fiber supplementation during gestation or transition periods improves sow reproductive performance by reducing low-viable and stillborn piglets, diarrhea and mortality, and enhancing colostrum production. High fibre intake alters gut microbiota composition and function, although physiological mechanisms on host remain unclear. This study aimed to profile fecal microbiota and blood metabolome of eighty-eight Large White × Landrace sows assigned to 4 treatments including: high fiber (500g straw pellets) or low fiber (no straw pellets) diets; hessian (30×60cm piece of material) or straw (one biscuit of wheaten straw) enrichments from entry to farrowing (4.1 ± 0.1 days) in a 2×2 factorial arrangement before farrowing. Blood and fecal samples were collected two days postpartum and analysed using untargeted uHPLC-MSMS metabolomics and 16S rRNA amplicon sequencing technique, respectively. High fibre diet up-regulated tryptanthrine,

isoquinolines (anti-inflammatory metabolites), and glucose/energy homeostasis via bile salt production ($P < 0.001$), and downregulated indole-3-acetate (linked to pro-inflammatory cytokine inhibition; $P < 0.001$). No difference was observed for enrichment type. Beta diversity analysis of microbiota revealed significant differences in fecal microbiota between treatments. The main families affected in treatment groups included *Prevotellaceae, Acidaminococcaceae, Eubacterium coprostanoligenes group*. High-fibre/hessian treatment increased *Prevotellaceae* and *Acidaminococcaceae*, while *Eubacterium coprostanoligenes group* was more abundant in the hessian/low-fibre treatment. Significant differences between faecal microbiota of sows on high and low fibre diets were observed, although the enrichment type had no effect on faecal microbiota. In conclusion, high-fiber supplements before farrowing, even short-term, influences metabolic and microbial profiles, highlighting the role of microbial metabolites in the benefits of high fiber diets. **Key Words:** Sow, fibre, microbiota

**19 Yeast produced on milk permeate reduce post-weaning diarrhea in piglets.** N Canibe*, K Jerez-Bogota, KE Bach Knudsen, and SK Jensen, *Aarhus University, Department of Animal and Veterinary Sciences, Blichers Allé 20, 8830 Tjele, Denmark*. Lactose-rich permeate is a side stream from the production of various dairy products. Components of yeast cells, i.e., α-D-mannans and β-D-glucans, have shown beneficial effects on health. The aim of the study was to investigate the preventive effect of a permeate-grown yeast product on post-weaning diarrhea and related parameters in an enterotoxigenic *E. coli* **(**ETEC) F18 challenge study. Two yeast strains were grown on permeate with urea added as nitrogen source. The produced yeast biomass was concentrated and subsequently spray dried. Sixty weaners (~28d) from 10 sows confirmed homozygous carriers of the FUT1 gene were used. The pigs were assigned to one of three groups (20 pigs per group; two per pen): Non-challenge control (N-Ch); challenge control (Ch); and challenge fed the control diet supplemented with 2.5% yeast product (Yeast). Pigs were fed the diets for three weeks from weaning. The challenge groups were inoculated with $1x10^{10}$ cfu ETEC F18 on d8 and 9 postweaning. Fecal samples (10 per pig) were collected for consistency score, DM determination and quantification of ETEC F18 fimbria by qPCR. Blood was collected (6 samples per pig) for haptoglobin and PigMAP analysis. Fecal DM% was lower ($P < 0.05$) in the Ch than in the N-Ch group during the study duration, with the Yeast group showing intermediate values but not different from the other two groups. The highest diarrhea incidence was measured on d4 to 6 post-first challenge day, with lower values for the Yeast than Ch on both days, i.e., d4 ($P < 0.05$) and 6 ($P < 0.10$). Coinciding with the highest fecal score and lowest DM%, fecal shedding of ETEC F18 fimbria was highest on d4 and 6 post-first challenge day in both challenged groups, but with lower copy numbers in the Yeast than the Ch on both days ($P < 0.01$). Haptoglobin and PigMAP concentrations were not affected by challenge or yeast supplementation. In conclusion, the yeast product was able to reduce diarrhea in ETEC F18 challenged pigs. **Key Words:** post-weaning diarrhoea, side stream, yeast

**20 Stimulation of bacterial growth from porcine strains by neuroendocrine catecholamines.** J d'Amedor de Mollans, F Blanchet, and D Guillou*, *Mixscience, Bruz, Bretagne, France*. Avoiding stress in a pig's productive life is impossible, and stressful events are linked to health issues like scours or infection. Microbial endocrinology examines how animal hormones interact with gut bacteria, with pathogens responding more than beneficial commensals. A series of 3 trials were conducted to transpose methods from the literature to the lab and to set up internal references of the effect of neuroendocrine catecholamines: dopamine (DOPA), epinephrine (EPIN), norepinephrine (NORE), and of cortisol (CORT) on bacterial growth *in vitro*. Strains of *Staphylococcus aureus* (SA), *Streptococcus suis* (SS) and *Escherichia coli* (EC) from the lab collection were grown in a broth of minimal nutrient medium (SAPI) complemented with adult bovine serum to mimic intestinal content, or on nutrient rich medium (BHE-YE) as positive control. Bacterial culture growth was studied using a continuous method (microplate spectrophotometry), confirmed by end-point plate counting then analyzed by a one-way Student test. To select hormone concentration, optical density of SA culture was measured over 72 hours with hormones concentrations of $10^{-4}$, $10^{-3}$ and $10^{-2}$ mol/L for a starter inoculum set at $10^{1}$ CFU/mL. NORE concentration of $10^{-3}$ mol/L stimulated SA growth, but

with a $10^{-2}$ mol/L concentration, growth time lag and log phase were shortened. To observe bacterial growth in all media and species, $10^2$ UFC/mL of inoculum was required. All hormones decreased significantly the latency of the exponential phase of SA ($P < 0.05$). NORE and DOPA decreased latency of the exponential phase in SS and EC or with SS, they also boosted generation times ($P < 0.05$). To conclude, stress hormones boosted pathogenic bacteria growth, especially in the exponential phase. Because the effect depended on hormonal concentration and inoculum density, further research should be undertaken to collect relevant values observed in the pig intestine. **Key Words:** Stress hormones, pathogenic bacteria, in vitro model

**21 Analysis of cecal microbial communities in weaned pigs fed high canola meal diet without or with acidifier and their association with glucosinolate metabolism.** Emily Fowler*[1], Jinsu Hong[2], Crystal Levesque[1], and Benoit St-Pierre[1], [1]*Department of Animal Science, South Dakota State University, Brookings, SD, USA*, [2]*Department of Animal Science, University of Minnesota, Saint Paul, MN, USA*. Glucosinolates present in canola byproducts limit their incorporation into swine diets due to effects on the thyroid and feed intake, leading to growth retardation. As the majority of microbial glucosinolate metabolism occurs in the hindgut, investigating bacterial communities in this compartment may yield valuable insights. To this end, this study identified the major cecal bacterial species of weaned pigs fed canola meal (CM)-containing diets without or with feed additive to reduce gut pH. Cecal digesta samples were collected from 40 weaned pigs fed five experimental diets for 5 weeks: CM 20% diet, CM 40% diets without or with acidifier or butyrate or both acidifier and butyrate. Samples were analyzed using the 16S rRNA gene through PCR-amplification of the V1-V3 regions from microbial genomic DNA, followed by Illumina MiSeq 2X300 sequencing. Comparisons of abundance for OTUs amongst dietary treatments were performed in R using the non-parametric Kruskal–Wallis test, followed by the Wilcoxon test for multiple pairwise comparisons. While no significant differences in composition were found among diets, two OTUs [Ssd-00001, *Lactobacillus amylovorus* (99.64%); Ssd-00039 (*Streptococcus macedonicus*; 94.75%)], were most abundant overall. In metagenomic analysis, an average of 498 contigs were generated for a combined length of 10,056,793 bp, which included an average of 16,075 coding sequences. Predicted metabolic functions included pathways involved in carbohydrate utilization (glycolysis) and production of lactic acid and short-chain fatty acids. Notably, eight candidate homologs of a glycoside hydrolase GH1 family enzyme (EC3.2.1.86) previously reported to be responsible for the degradation of sinigrin, a major glucosinolate present in the Brassicaceae family, were identified. Together, these findings provide insights into the metabolic capabilities of cecal bacterial species in pigs fed a CM-based diet, including their potential for breaking down glucosinolates. **Key Words:** canola meal, glucosinolate, pig

**22 Impact of in-feed fucosidase on pig gut microbiota and health.** D. Georgaki*[1], O. Højberg[1], A.A Schönherz[1], C. Poulsen[2], and N. Canibe[1], [1]*Animal and Veterinary Sciences, Aarhus University, Denmark*, [2]*IFF International flavors and fragrances, Denmark*. Weaning is a critical period that introduces multiple stressors, leading to e.g., piglet postweaning (**PW**) diarrhea or longer lasting gut dysfunctions. Here, we aimed to study the potential role of in-feed 1,2-α-L-fucosidase in modulating pig gut microbiota. This exoglycosidase facilitates the fucose release, a key component of pig gut mucins and plant polysaccharides, for microbial utilization, and is hypothesized to improve gut health by promoting beneficial bacteria, like propionate or butyrate producers. Weaned piglets (n=240) were divided into four groups: a control and three fucosidase treatments, receiving fucosidase (100 mg/kg feed) for 2-, 7-, or 16-weeks PW, respectively. Gut digesta and tissue from small intestine and colon were collected (d10, 43, and 114 PW) for microbiota (qPCR, 16S rRNA sequencing) and gene expression analysis, respectively. Gut epithelium tissue was tested for MUC1, MUC2, FUT1, FUT2 gene expression, relative to a reference gene (UBC). Further, feces and blood samples were collected (d1, 5, 10, 43, 72, 100 PW) for dry matter analysis and hematology parameters, respectively. Microbiota composition was assessed using principal response curves, alpha diversity metrics (richness and Shannon index), and differential abundance analysis (DESeq2). Mixed effects models were used. Alpha

diversity (p≥0.38) and overall microbiota composition (p≥0.09) did not differ significantly between treatment groups. In distal colon on d114 PW, members of *Roseburia* and *Blautia*, known propionate producers, increased ($P < 0.001$) in the enzyme supplemented treatment groups compared to control group. The abundance of *Prevotella* genus members was also affected by enzyme addition (p≤0.05) but showed individual and contrasting patterns. The enzyme did not affect host gene expression, blood parameters, and fecal dry matter. Thus, our findings demonstrated in-feed fucosidase to modulate the pig gut microbiota, with no clear conclusion regarding gut or host health. **Key Words:** Fucosidase, Gut microbiome, Gut health

**23 Inclusion of spray dried porcine plasma as a zinc oxide alternative in creep feed and pre-starter diets of piglets impacts plasma metabolites and gut microbiota.** Z.W. Ng'ang'a*[1,2], J. Tarradas[1], N. Tous[1], R. Beltrán-Debón[2], P. Javier[3], L. Laghi[4], F. Correa[5], D. Luise[5], P. Trevisi[5], and D. Torrallardona[1], [1]*IRTA, Animal Nutrition, Constantí, Catalonia, Spain*, [2]*MobioFood Research Group, Universitat Rovira i Virgili, Tarragona, Catalonia, Spain*, [3]*APC Europe, S.L., Granollers, Barcelona, Spain*, [4]*Department of Agricultural and Food Science, University of Bologna, Cesena, Italy*, [5]*Department of Agricultural and Food Science, University of Bologna, Bologna, Italy*. We evaluated spray-dried porcine plasma as an alternative to zinc oxide in weaned piglets, focusing on performance, plasma metabolomics and gut microbiota. At 7d of age, 23 litters (mixed sex) were offered one of 3 creep feed treatments: Control (CO), ZnO (3%) or SDPP (5%). From weaning to 14d PW, a selection of 72 piglets per group were distributed into 36 pens according to a RCBD (6 pigs/pen; mixed sexes; 12 blocks of BW). ZnO and SDPP improved ADG and ADFI compared to CO (0-14d PW; $P < 0.05$). At 14d PW, 2 piglets per pen were used to obtain plasma for H-NMR spectroscopy, and jejunal and cecal content for 16S rRNA gene sequencing. The metabolomic analysis showed that CO had higher β-Ala and X2-aminobutyrate, ZnO increased glucose, Gly, Met, Val and Ile while SDPP had increased Tyr, Thr, ethanol and sarcosine ($P < 0.05$). Diet affected β-diversity in caecum ($R^2 = 0.07$, $P = 0.001$) and in jejunum ($R^2 = 0.05$, $P = 0.08$). CO had higher abundance of *HT002* in jejunum and *Campylobacter* in caecum (LDA > 4, $P < 0.05$). ZnO was characterized by *Terrisporobacter* and *Turicibacter* in both jejunum and caecum (LDA > 3.9, $P < 0.05$) as well as *Clostridium sensu stricto 1* and *Prevotella* in caecum (LDA > 4.2, $P < 0.05$). SDPP was characterized by *Megasphaera*, *Limosilactobacillus* and *Actinobacillus* in caecum (LDA > 3, $P < 0.05$), while *Sarcina*, *Actinobacillus*, and *Romboutsia* had the highest abundance in jejunum (LDA > 3, $P < 0.05$). Multiblock sPLS-DA correlation analysis showed significant relationships between metabolites and microbial groups (i.e. betaine and Met with *Clostridium sensu stricto* 6 and Ile with *Terrisporobacter;* but only in ZnO), suggesting diet-specific influences. Overall, SDPP promoted growth performance of piglets similarly to ZnO. However, results on the metabolite and microbial analysis suggested different modes of action between SDPP and ZnO. Therefore, SDPP may be used instead of ZnO to support gut health and performance of weaned piglets. **Key Words:** Gut health, weaning, microbial metabolism

**24 The effect of protein level and essential amino acid supplementation on the gut microbiota and its metabolic function in weaned piglets.** M. H. Kroier[1], A. A. Schönherz[1], H. N. Lærke[1], N. M. Sloth[2], M. Loomans[3], J. J. Koehorst[3], M. Suarez-Diez[3], and N. Canibe*[1], [1]*Department of Animal and Veterinary Sciences, Aarhus University, Foulum, Blichers Allé 20, DK-8830 Tjele, Denmark*, [2]*SEGES innovation P/S, Agro Food Park 15, DK-8200 Aarhus N, Denmark*, [3]*Laboratory of Systems and Synthetic Biology, Wageningen University & Research, Stippeneng 4, 6708 WE Wageningen, The Netherlands*. We studied how protein levels and extra essential amino acids (EAAs) supplementation influence gut microbiota composition and function, focusing on their potential to mitigate the adverse effects of a high-protein diet. A total of 120 weaners (~28d old) were fed one of three diets: Lp (15% crude protein (CP)), Hp (25% CP), and HpEAA (Hp with 35% extra crystalline lysine above recommendations, and corresponding increases in methionine, threonine, tryptophan, and valine of 32, 62, 22, and 67% relative to lysine). Pigs were fed ad libitum for 4 weeks. Feces from each pen were scored for diarrhea daily. On d7 and 28 post-weaning, 20 pigs per diet were euthanized, and gut digesta and mucus sampled from distal small intestine (SI3), cecum

(CAE), proximal (CO1), mid (CO2), and distal (CO3) colon for microbial composition (16S and ITS sequencing) and microbial metabolite analysis. Linear mixed-effects models were used to assess statistical differences. From week 2, pigs fed Lp had lower diarrhea odds than those fed Hp and HpEAA ($P < 0.05$); and in week 3, HpEAA had lower odds than Hp ($P < 0.05$). On d7, Lp pigs had greater fungal alpha diversity than Hp ($P < 0.05$) and HpEAA ($P < 0.05$) in CO1 and CO2. All diets had different bacterial composition (Bray Curtis) in mucus ($P < 0.05$) and CAE digesta ($P < 0.05$); whereas in SI3, CO1, and CO2, only Lp differed from Hp ($P < 0.05$) and HpEAA ($P < 0.05$). On d28, *Limolactobacillus* and *Lactobacillus* were more abundant in SI3 of HpEAA pigs than in Lp ($P < 0.05$). HpEAA tended to have lower SCFA concentration than Hp ($P < 0.05$) in colon. In CAE and colon, Hp had generally higher ($P < 0.05$) levels of indole, skatole, and p-cresol than Lp, with HpEAA showing intermediate values. Total biogenic amines tended to be higher in HpEAA pigs ($P < 0.10$) than in Lp and Hp across segments. Extra EAA supplementation reduces diarrhea; however, the observed changes in gut microbiota and metabolites do not decipher its underlying mechanisms, warranting further data exploration. **Key Words:** Weaning, diarrhea, diet composition

**25 Effects of Sucrosomial® Iron on gut microbiota in term and preterm piglets.** X. Wang[1], P. Lipinski[1], M. Ogluszka[2], R. Mazgaj[1], J. Wolinski*[3,4], D. Szkopek[3], K. Zaworski[4], Z. Kopec[1], B. Zelazowska[1], G. Tarantinoe[5], E. Brilli[5], and R.R. Starzynski[1], [1]*Laboratory of Iron Molecular Biology, Department of Molecular Biology, Institute of Genetics and Animal Biotechnology, Polish Academy of Sciences, 05-552 Jastrzebiec, Poland*, [2]*Department of Genomics and Biodiversity, Institute of Genetics and Animal Biotechnology, Polish Academy of Sciences, 05-552 Jastrzebiec, Poland*, [3]*Laboratory of Large Animal Models, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, 05-110 Jablonna, Poland*, [4]*Department of Animal Physiology, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, 05-110 Jablonna, Poland*, [5]*Scientific Department, Pharmanutra S.p.A, 56122 Pisa, Italy*. Iron is essential for the healthy development of both the host and intestinal microbiota. However, intestinal immaturity in newborns may impair the effective utilization of iron. This study aimed to evaluate the effects of a novel iron supplement, Sucrosomial® Iron (SI), on newborn intestinal health. Twelve preterm piglets were born by cesarean section on the $109_{th}$ day of gestation from 3 Polish Landrace sows. As control, twelve term piglets born vaginally (115 days gestation) from 3 sows were obtained. Piglets were allotted into 4 groups: term and preterm piglets without or with SI (2 mg Fe/piglet/day) from day 4 to day 10. Samples were collected on day 11. Data were analyzed using ANOVA (general linear model) followed by Duncan's multiple comparison tests for post hoc analysis. Premature delivery led to a lower duodenal villus height: crypt depth ratio than vaginal delivery ($P < 0.05$). Low levels of key factors involved in duodenal iron absorption were observed in preterm piglets ($P < 0.05$). In addition, preterm piglets exhibited differences from term piglets in alpha and beta diversity, as well as in bacterial composition in the rectum ($P < 0.05$). SI improved duodenal morphology in both term and preterm piglets, and increased the protein levels of ferroportin and ferritin, while decreasing divalent metal transporter 1 levels in term piglets ($P < 0.05$). Beta diversity analysis revealed clear differences in microbiota composition between preterm groups fed with or without SI ($P < 0.05$). SI promoted the growth of beneficial bacteria *Ligilactobacillus* and *Limosilactobacillus*, and reduced the abundance of harmful bacteria *Clostridium sensu stricto* and *Bacteroides* in preterm piglets ($P < 0.05$). These results indicate differences between term and preterm piglets in iron absorption mechanisms and microbiota composition, and suggest that SI has a favorable effect on intestinal health. This study was supported by the National Science Centre, Poland (2020/39/B/NZ5/02469). **Key Words:** Sucrosomial® Iron, preterm piglet, intestinal microbiota

**26 Effects of a novel source of Magnesium-protected Zinc in controlling in vitro *Escherichia coli* proliferation.** Z. Garlatti*[1], V. Courtois[2], E. Bacou[1], N. Joguet[2], T. Chalvon-Demersay[2], J. Le Cour Grandmaison[1], and A. Juanchich[2], [1]*TIMAB Magnesium, Dinard, France*, [2]*Centre Mondial de l'Innovation Roullier, Saint-Malo, France*. Post-weaning diarrhea (PWD) remains a major challenge in piglet health management, often caused by pathogenic strains of *Escherichia coli* (*E. coli*). Traditionally, high-dose zinc oxide (ZnO) has been used to control PWD due to its antimicrobial properties. However, environmental concerns over zinc pollution and antimicrobial-resistance spread have prompted the search for alternatives. The study evaluated the effect of a novel source of Magnesium-protected Zinc (Mg-Zn) compared to ZnO on the in vitro proliferation of *E. coli*. The protocol implemented is referred to as an "on-spot" protocol: the products to be tested are incorporated into the nutrient agar, and *E. coli* (DSMZ 682 strain) proliferation is assessed by measuring the size of the colonies at days 1 and 7 post-deposition of the bacteria on the agar. All conditions were tested in 3 replicates. Two sources of Zn were evaluated, ZnO and Mg-Zn, at 100, 300, 750, and 2500 ppm Zn. A condition with no Zn added consisted of the control group. The results are analyzed using a two-factor ANOVA test (product and dose) to determine statistical significance ($P < 0.05$). The study shows that Mg-Zn significantly inhibited *E. coli* proliferation at 100 ppm Zn compared to the control group ($P < 0.001$), and at higher doses (750 or 2500 ppm Zn) both treatments achieved comparable inhibition. The atomic protection of Zn by Mg likely enhances its antimicrobial efficacy, offering a potential of growth inhibition from 100 ppm Zn with no statistical difference with a level of 2500 ppm Zn. Mg-Zn effectively controls *E. coli* growth at significantly lower Zn concentrations compared to ZnO. By addressing post-weaning diarrhea with reduced Zn levels, Mg-Zn represents a viable alternative to conventional high-dose ZnO. Future work will focus on field trials, testing Mg-Zn against diverse pathogens, and optimizing its integration into piglet feed to support healthier and environmentally sustainable weaning practices. **Key Words:** zinc, intestinal health, post-weaning diarrhea

**27 Exploring the prebiotic potential of non-digestible carbohydrates: insights from *In Vitro* fermentation pattern and microbial community.** N Razmgah*, N Canibe, A. A Schönherz, É Chassé, M Skou Hedemann, and K. E Bach Knudsen, *Department of Animal and Veterinary Sciences, Aarhus University, 8830 Tjele, Denmark*. Monomer composition and structure of non-digestible carbohydrates (NDC) impact microbial fermentability, lead to distinct microbial and metabolite profiles. A subgroup of NDC, i.e., prebiotics, selectively promote the growth of beneficial species. We aimed to study the fermentation and microbial profile of NDC sources differing in monomer composition and degree of polymerization. The study included arabinoxylan (AX), xylo-oligosaccharide (XOS), inulin (INU), fructo-oligosaccharide (FOS), fucoidan (FUC), chitosan (CHI), and chito-oligosaccharide (COS). *In vitro* fermentations were conducted by incubating a medium with 10% fecal slurry (Control, CON) at 37°C for 48 hours (h), alongside the CON medium supplemented with 2% of each of the substrates. Samples were collected at 0, 2, 6, 12, 24, and 48h to assess NDC degradation and short-chain fatty acid (SCFA) production. Bacterial counts (qPCR) and microbial profiling (16S rRNA sequencing) were evaluated at 0 and 48h. NDC degradation was between 65-98% for AX, XOS, INU, and FOS, being highest for INU and FOS ($P < 0.05$), whereas FUC was not degraded. This reflected in the SCFA production, being significantly higher ($P < 0.05$) for the most degradable compared to the least degradable substrates. At 48h, alpha diversity (Shannon index) of all substrates except COS and FUC, which was not affected, was lower than that of the CON ($P < 0.05$). Principal Coordinate Analysis revealed microbial composition differences between substrates (PERMANOVA, $P < 0.001$), forming 3 clusters: AX, XOS, FOS, and INU; CHI and CON; and COS. From a heatmap, *Lactobacillus* and *Prevotella* abundance increased at 48h in AX, XOS, FOS, and INU, while FUC resulted in increased abundance of *Lactobacillus*. COS increased the abundance of *Clostridium*. In conclusion, the structures of NDC influenced its degradation, SCFA production, and microbiota composition, with AX, XOS, INU and FOS having similar patterns, COS a distinct pattern, and FUC and CHI showing subtle effects. **Key Words:** prebiotic, fermentation, microbiota

**28 Functional ingredients to optimize gut functionality in post weaning piglets.** S. K. Kar, E. Zaccaria, G. Binnendijk, P. van Wikselaar, and A. J. M. Jansman*, *Wageningen Livestock Research, De Elst 1, 6708 WD Wageningen, The Netherlands*. The effects of functional ingredients in diets for post-weaning (PW) piglets on performance and functionality of the gut were studied using six dietary treatments: inclusion of 5% alfalfa meal (non-fermentable fibre) or 5% beet pulp (fermentable fibre), as plant based fibre diets (PFD), or 3% black soldier fly (BSF) insect oil or 5% BSF chitin-rich meal, as insect-based diets (IBD) and a control and reference diet supplemented with 3000 mg Zn as ZnO/kg. Diets were nutrient and energy balanced and provided for 13 d PW. Each treatment was evaluated in 12 replicates (floor pens with 10 piglets each). In the next three weeks a common phase II diet was provided in all treatments. Samples of blood, intestinal tissue and digesta, and faeces were collected from one piglet per pen on d 13 and 34 PW to assess inflammatory markers (cytokines, acute-phase proteins), faecal consistency, transcriptomics in ileal tissue, ileum and colon microbiome composition, and short-chain fatty acid (SCFA) concentrations in colonic digesta. Data were statistically analysed using univariate and multivariate methods. The inclusion of BSF chitin-rich meal significantly ($P < 0.05$) enhanced ileal microbiome alpha-diversity indices, highlighting its potential to support microbial stability in the gut. Despite small numeric improvements in faecal consistency during the first 13 days PW, inclusion of PFD and IBD did not affect growth performance, health indices, transcriptomic profiles in ileum tissue, or digesta SCFA concentrations compared to the control diet. In contrast, ZnO supplementation significantly ($P < 0.05$) improved growth performance, modulated gene expression, enhanced colonic microbiome diversity, SCFA profiles, and suppressed *E. coli*. These effects, however, did not sustain throughout the entire experimental period. In conclusion, selected plant- and insect-based ingredients showed limited effects in relation to gut health and functionality in the immediate PW phase. **Key words:** Functional ingredients, Gut health, Intestinal microbiome

**29 Influence of corn based fibrous co-products on colon mucosa gene expression and mucosal- associated microbiome of growing pigs.** H. Miller*[1], C. Anderson[2], S.S. Schmitz-Esser[2], A. Ericsson[1], and A.L. Petry[1], [1]*University of Missouri, Columbia, MO, USA*, [2]*Iowa State University, Ames, IA, USA*. The experimental objective was to determine the influence of insoluble corn-based fibers (ICBF) on the colon microbial communities and transcriptome of growing pigs. Two replicates of 28 gilts (26.7±2.5 kg BW; PIC800 x Camborough; N = 56), were randomly assigned to 1 of 7 semi-synthetic diets. Dietary treatments included a control (CTL) diet with a formulated total dietary fiber (TDF) value of <1% and 6 diets with an ICBF replacing 30% of the corn starch in CTL: dehulled degermed corn (DHDG; TDF = 0.9%), ground corn (COR; TDF = 3.8%), corn gluten meal (CGM; TDF = 4.4%), high protein dried distillers grains (HP; TDF = 7.0%), dried distillers grains (DDGS; TDF = 7.9%), and corn bran (BRN; TDF = 12.3%). Pigs were individually housed and limit-fed 2.4 times maintenance. On day 31 pigs were necropsied. Mucosal scrapings and tissue were collected from the colon for 16S rRNA microbiota analysis and host differential expressed genes (DEG), respectively. Individual operational taxonomic units (OTUs) were compared using Linear Discriminant Analysis Effect Size. Data for alpha diversity metrics were analyzed as a mixed model with replicate as random effect and treatment as fixed effect. Eighty-six of the top 100 OTUs differed across dietary treatments ($Q < 0.05$). Observed species richness and Chao1 alpha diversity index increased with the inclusion of ICBF, while BRN was reduced relative to the other high fiber diets ($P < 0.05$). Diets containing ICBF were compared to CTL for DEG. Protein-coding genes were selected based on a false discovery rate of rate ≤0.05 and log2 fold change >1. Of the 160 genes that met the criteria, 140 came from either HP, DDGS, or BRN. MUC2 and MUC5AC, genes associated with mucin formation, were up regulated in BRN relative to CTL ($Q < 0.05$). LEAP2, an antimicrobial peptide, was decreased in DDGS relative to CTL ($Q < 0.05$). Collectively, these data show how changes in ICBF in the diet can alter mucosal microbiota and the associated gene-expression of the host. **Key Words:** colon, fiber, mucin

**30 Review of the effect of *Saccharomyces cerevisiae* supplementation in sows on reproduction performance under commercial conditions.** O. Merdy[1], H. Legendre[1], T. Kiros*[1], and F. Machuron[2], [1]*Phileo by Lesaffre, Marcq-en-Baroeul, France*, [2]*Lesaffre Institute of Science and Technology, Marcq-en-Baroeul, France*. The yeast probiotic *Saccharomyces cerevisiae* NCYC Sc47 (YP) has been shown to provide health benefits by modulating the pig microbiota, helping improve feed digestibility, controlling pathogen pressure, and offering immunomodulatory benefits. This study aimed to meta-analyze the recent field experience with YP supplementation of sows during late gestation and through lactation on reproductive performance under commercial conditions. The review was performed in Phileo by Lesaffre trial central database. Both field randomized trials and historical data records (records collected before and after introduction of YP) were selected. The statistical model included the fixed effect of the treatment (YP or control), the random effect of the trial and the block effect of the breeding cycle. Size effect was calculated by mean difference with 95% confidence interval. Heterogeneity among studies was estimated by $I^2$ value. The studies which drive a significant part of overall heterogeneity were identified by funnel plot and Baujat's plot analysis and then were excluded. The statistical analysis was conducted using the R package meta from R software (v4.0.3). From 9 trials inventoried, 2 of them were excluded due to respectively the absence of variability indicators and the use of YP at a different dosage. There were 6 randomized trials (30% of observations) and 1 was a historical data record. Two of the randomized trials were performed on 2 consecutive breeding cycles of the same sows. The trials involved more than 3,000 weaned litters. The meta-analysis showed that administering YP to sows resulted in an increase of 0.30 piglets weaned per litter ($P < 0.001$). YP supplementation also led to 230g higher individual bodyweight ($P < 0.001$) and 4.12 kg heavier litters at weaning ($P < 0.001$). In conclusion, supplementing sow diets with yeast probiotic *Saccharomyces cerevisiae* from the end of the gestation period increased the reproductive performance of sows. **Key Words:** yeast probiotic

**31 The impact of DDGS withdrawal in diets with and without an ionophore on gene expression and pathway activation in the ileum in late finishing pigs.** K.L. Saddoris-Clemons*, K.J. Bolek, and B.D Humphrey, *Phibro Animal Health, Teaneck, NJ, USA*. DDGS and ionophores are a common feed ingredient and additive utilized during the grower-finisher period in the USA. Limited information exists on the impact of reducing DDGS levels in the late finisher period on the GIT. A total of 72 grow-finish pigs (Initial BW = 75.1 ± 5.9 kg, 2 pigs/pen) were utilized to determine the impact of removing 30% DDGS from diets with and without an ionophore on gene expression and pathway activation in the ileum. Dietary treatments consisted of 1) 30% DDGS for 42d, 2) 30% DDGS for 35d followed by 0% DDGS for 7d, and 3) Trt 2 +18.1 g/ton narasin for the full 42d. Body weights were collected weekly and pen feed intake measured. On d42, all pigs were euthanized and mucosal scrapings were collected from the ileum. RNA was isolated from the mucosa and mRNA was sequenced for transcriptome and metagenomic analysis to identify differentially expressed genes and pathway activation. Feed conversion was worse ($P < 0.05$) in pigs fed Trt 3 compared to pigs fed Trt 1, and intermediate to pigs fed Trt 2. Pigs fed Trt 2 had a reduction ($P < 0.05$) in pathways involving nutrient absorption and metabolism, especially fats, compared to Trt 1. Several immune activation and oxidative pathways stress pathways were inhibited ($P < 0.05$), however cellular stress was activated ($P < 0.05$) in pigs fed Trt 2 compared to Trt 1. Pigs fed Trt 3 had reductions ($P < 0.05$) in pathways involved in nutrient absorption and metabolism, oxidative stress, and innate immune activation compared to Trt 2. Pathways involved in production of pro-inflammatory cytokines, vit E synthesis, and heat stress, were activated ($P < 0.05$) in pigs fed Trt 3 compared to pigs fed Trt 2. Withdrawal of DDGS impacted feed efficiency and ileal transcriptome pathways related to immune suppression, cellular stress, and absorption and metabolism of nutrients. Narasin reduced feed efficiency upon DDGS removal and exacerbated the negative effects on ileal transcriptome pathways. **Key Words:** pigs intestine DDGS

**32 The impact of DDGS withdrawal in diets with and without an ionophore on microbial composition and function in the intestine of late finishing pigs.** K.L. Saddoris-Clemons*, K.J. Bolek, and B.D Humphrey, *Phibro Animal Health, Teaneck, NJ, USA.* A total of 72 grow-finish pigs (Initial BW = 75.1 ± 5.9 kg) were utilized to determine the impact of removing 30% DDGS from the diets with or without an ionophore on microbiome composition and function. Barrows were randomly allocated to one of three dietary treatments: 1) 30% DDGS for 42d, 2) 30% DDGS for 35d followed by 0% DDGS for 7d, and 3) Trt 2 + 18.1 g/ton narasin for the full 42d. Individual fecal grab samples were collected on d35 and d42 for SCFA analysis and all pigs were euthanized and intestinal contents collected from the ileum and cecum on d42. DNA was extracted from ileal, cecal and d42 fecal samples, and analyzed by metagenomic shotgun sequencing. The relative abundance of microbial genera and species and functional clusters were modeled using the ANCOM-BC package in R. Pigs fed T3 tended ($P < 0.10$) to have higher amounts of lactic acid in the feces on d42 compared to pigs fed T2. Pigs fed T2 and T3 had reduced ($P < 0.01$) alpha-diversity in the ileum, but greater ($P < 0.05$) alpha-diversity in fecal samples compared to pigs fed T1. Pigs fed T2 has increased ($P < 0.001$) abundance of *Aequoribater fuscus* in the ileum and reduced ($P < 0.001$) *Ruminococcus* and *Prevotella* species in feces compared to pigs fed T1. Several (50+) Level 3 KEGG pathways increased in abundance ($P < 0.01$) in the cecum of pigs fed T2 compared to pigs fed T1. In feces, KEGG pathways related to starch metabolism decreased ($P < 0.01$) while pathways involved in fatty acid and lysine metabolism increased ($P < 0.01$) in relative abundance in pigs fed T2 compared to pigs fed T1. Narasin increased ($P < 0.001$) the genera of *Nocardiopsis* and *Streptomyces* and increased relative abundance ($P < 0.01$) of genes related to polyketide biosynthesis in the ileum. Removal of DDGS increased the genetic representation of metabolic pathways in the lower GIT. Narasin impacted microbial composition in the ileum, but increased lactate in the feces following the removal of DDGS. **Key Words:** pigs microbiome DDGS

**33 F18 *E. coli* impacts intestinal secretion but not barrier function in a weanling pig model.** S.C. Pearce*[1], M.J. Nisley[2], E. Due[2], E.R. Burrough[3], and N.K. Gabler[2], [1]*USDA-ARS, Ames, IA, USA,* [2]*Department of Animal Science, Iowa State University, Ames, IA, USA,* [3]*Vet Diagnostic & Production Animal Medicine, Iowa State University, Ames, IA, USA.* F18 Enterotoxigenic *Escherichia coli* (ETEC) binds to the intestinal epithelium, induces post-weaning diarrhea (PWD) and edema disease, and compromises intestinal integrity in weaned pigs. Our objective was to evaluate the extent to which F18 ETEC disrupts ileum epithelial barrier integrity during the early onset of disease. 96 pigs (5.4 ± 0.92 kg body weight) were allocated by sex and FUT1 genotype (gene that can influence ETEC attachment) across 16 pens (6 pigs/pen) in multiple rooms and fed a common diet. After 10 days on diets, half the pigs were orally administered 5 mL of $3.8\times10^9$ cfu F18 ETEC *E. coli* culture (day post-infection [dpi] 0). On dpi 5, one pig/pen was sacrificed to assess ileal morphology and barrier forming tight junctional protein claudin 3 (CLDN3), pore-forming tight junctional protein claudin 2 (CLDN2), and anion channel protein cystic-fibrosis transmembrane conductance regulator (CFTR) protein via immunohistochemistry. Fresh ileal tissues were assessed in Ussing chambers for the macromolecular flux of FITC-Dextran (FD4), transepithelial electrical resistance (TEER), and nutrient flux. At dpi5, there were no differences in ileum FD4 or TEER. Similarly, there were no differences between CON and ETEC in nutrient flux of glutamine or glucose. Ileum pH was significantly increased ($P < 0.01$) from 6.08 to 7.10 in ETEC-challenged pigs. Barrier-forming protein CLDN3 was significantly decreased in ETEC-infected pigs by 15% ($P < 0.001$). Similarly, pore-forming paracellular cation transport protein CLDN2 was decreased 24% in ETEC-infected pigs compared to CON ($P < 0.05$). Interestingly, chloride secretion protein CFTR was increased 85% in ETEC-infected pigs compared to CON ($P < 0.001$). CFTR impacts bicarbonate secretion which may result in increased luminal pH. In conclusion, at dpi 5, intestinal barrier function may not be severely impacted, but proteins involved in the transport of molecules across the intestinal barrier are altered, including ion transport. **Key Words:** pathogen, barrier function, transport

**34 Impact of postbiotics, probiotics and plant extract feed additives on physiological responses and microbiome of gilts exposed to heat and farrowing stressors, and their offspring microbiome.** H Hedrick[1], A KNOELL[2], T Safranski[1], A Petry[1], M Lucy[1], C González-Vega[3], M Sardi[2], A Naqvi[4], M Le Gall*[3], and E Khafipour[3], [1]*Division of Animal Science University of Missouri, Colombia, MO, USA*, [2]*Core R&D Cargill Inc, Minneapolis, MN, USA*, [3]*Animal Nutrition and Health, Cargill Inc, Minneapolis, MN, USA*, [4]*Data Science and Engineering Cargill Inc, Minneapolis, MN, USA*. The objective was to evaluate the effects of three feed additives under gestation heat stress (HS) and farrowing stress (FS) on physiological responses and microbiome. At d 70 of gestation, Large White x Landrace gilts were allotted and fed one of four dietary treatments (n = 12/treatment) until the end of lactation: a control (CON) diet; a diet with 0.2% combination of a *Saccharomyces cerevisiae* postbiotic and a Bacillus probiotic prototype (SCB); a diet with 0.025% of an essential oil blend mixed with spices from the Fabaceae family and a polyphenol enriched plant extract (EO); or a diet with 0.2% *S. cerevisiae* postbiotic (SCFP). The room temperature cycled daily between 28-38°C from d 95 to 111 of gestation and reduced to ~22°C for the rest of study. Fecal samples were collected from gilts on d 70, 90, and 111 of gestation, and d 1, 4 and 21 of lactation, and from piglets (n = 72/treatment) at weaning and subjected to shotgun metagenomics. Data were analyzed using PROC MIXED of SAS, V9.4, with treatment as fixed and rep as random factors. For repeated observations, time and treatment X time were added in the model. Rectal temperature and breaths per min (BPM) increased ($P < 0.01$) with HS in all treatments; however, were lower ($P = 0.08$ and $< 0.01$, respectively) in SCFP (65.4 BPM, 38.3°C) gilts than EO (80.5 BPM, 38.5°C) and SCB (75.5 BPM, 38.5°C) gilts. Farrowing duration was longer ($P < 0.01$) in EO (5.6 hr) gilts than SCB (3.2 hr) and SCFP (3.1 hr) gilts. Disturbance in microbiome was observed during the transition from the gestation to lactation and post-farrow with the SCFP gilts having a stable and EO gilts the most decline in diversity ($\delta = 0.05$ vs. -0.67, $P < 0.05$). The piglet microbiome of SCFP fed sows had the most suppressive effects (0.70 vs. 0.38, 0.12, and 0.05, $P < 0.1$), reducing the ability of potential pathogens to compete. In conclusion, SCFP additive ameliorated the negative impacts of HS and FS and resulted in a more favorable piglet microbiome. **Key Words:** Phytogenic, Postbiotic, Probiotic

**35 Maternal supplementation with *Bacillus altitudinis* WIT588 improves porcine offspring growth performance and carcass weight and modulates ileal and faecal microbiota composition.** G. E. Gardiner[1], J. T. Cullen*[1,2], D. Crespo-Piazuelo[2], R. Rattigan[1], P. Cormican[3], and P. G. Lawlor[2], [1]*Eco-Innovation Research Centre, Department of Science, South East Technological University, Waterford, X91 K0EK, Ireland*, [2]*Pig Development Department, Animal and Grassland Research and Innovation Centre, Teagasc, Moorepark, Fermoy, Co. Cork, P61 C996, Ireland*, [3]*Animal and Bioscience Research Department, Animal and Grassland Research and Innovation Centre, Teagasc, Grange, Dunsany, Co. Meath, C15 PW93, Ireland*. The objective was to assess the effect of feeding *Bacillus altitudinis* to sows and/or offspring on growth, health indicators and microbiota. On day (D) 100 of gestation, sows were grouped as: control (CON; standard diet; n = 12 sows) and probiotic (PRO; standard diet supplemented with *B. altitudinis* WIT588 from D100 of gestation until weaning at ~26 days; n = 12 sows). At weaning, 72 piglets/sow treatment were selected, formed into same-gender pen groups of 2 pigs which were randomly assigned to either a CON (no probiotic) or PRO (*B. altitudinis*) treatment for 28 days post-weaning (pw), resulting in 4 treatment groups (n = 18 pens/treatment; maternal treatment/offspring treatment): 1) CON/CON; 2) CON/PRO; 3) PRO/CON; 4) PRO/PRO. Treatment allocation was balanced for gender. Data were analysed using the MIXED procedure in SAS for a 2 × 2 factorial arrangement except for OTU abundance which was analysed using DeSeq2 in R. *B. altitudinis* WIT588 was detected in the faeces of probiotic-supplemented sows and their piglets, and in the faeces and intestine of probiotic-supplemented piglets. Colostrum from PRO sows had a higher protein concentration ($P < 0.05$) and a higher relative abundance of *Rothia* ($P < 0.05$). Maternal treatment improved offspring feed conversion ratio at D0-14 pw ($P < 0.001$) and increased offspring BW at D105 and D127 pw ($P < 0.01$), carcass weight ($P < 0.05$) and kill-out yield ($P < 0.01$). It also increased offspring duodenal villous height at D8 pw ($P < 0.01$). Polysaccharide-fermenting (*Prevotella*) and butyrate-

producing (*Blautia*) bacteria were more abundant in the ileum of PRO/CON than CON/CON offspring on D8 pw ($P < 0.05$) and *Lactobacillus* was more abundant in PRO/CON faeces on D118 pw ($P < 0.01$). Overall, maternal, rather than pw, probiotic supplementation had the most impact, with improved colostrum quality, increased small intestinal villous height and ileal microbiota modulation likely contributing to the lifetime growth benefits in the offspring of supplemented sows. **Key Words:** Probiotic Sow Microbiota

**36 Dietary Iron Source Modulates Gut Microbiome Composition and Reduces Post-Weaning Diarrhea in Nursery Pigs Under Different Sanitary Conditions.** Sudario Roberto Silva Junior*[1], Maria Cristina Silva[2], Victor Hugo Silva Souza[4], Mamunur Rhaman[4], Lucas Rodrigues[3], Vinicius Cantarelli[2], Isabella Condotta[4], and Andres Gomez[1], [1]*University of Minnesota, Saint Paul, MN, USA*, [2]*Universidade Federal de Lavras, Lavras, MG, Brazil*, [3]*Zinpro Corpotation, Eden Praire, MN, USA*, [4]*University of Illinois Urbana-Champaign, Urbana, IL, USA*. This study aimed to investigate the effects of dietary Fe source on gut microbiome composition and diarrhea incidence in nursery pigs under different sanitary conditions. Weaned pigs (n=120; initial BW = 6.0 kg; 21 days old) were randomly assigned to four treatments in a 2 × 2 factorial design combining dietary Fe source [80 mg/kg Fe from either Ferrous Sulfate (FeSO4) or Ferric-Amino Acid complex (FeAA)] and sanitary conditions [4 pigs/pen housed in clean pens (CP) vs. 6 pigs/pen housed in dirty pens (DP)]. Diarrhea was scored daily (0-4 scale), and fecal samples collected on days 7 and 42 post-weaning. DNA was extracted from fecal samples and the V4 region of 16S rRNA bacterial gene was amplified and sequenced on the AVITI platform. Sequence data were processed to generate taxonomic abundances and different R packages were used to perform the analyses. Pigs fed FeAA had reduced diarrhea scores compared to FeSO4 at both days 7 ($P = 0.01$) and 42 ($P = 0.02$), regardless of sanitary conditions. Greater bacterial alpha diversity was observed in DP compared to CP pigs, regardless of Fe source fed at day 7 (Kruskal-Wallis, $P = 0.01$). Beta diversity, measured using Bray–Curtis, demonstrate that the iron source influences the gut microbiome depending on sanitary conditions (PERMANOVA, $P = 0.04$, $R^2 = 0.01$). Partial least squares discriminant analysis confirmed distinct clustering among treatments (AUC > 0.78). On day 7, pigs fed with FeAA under both sanitary conditions and those fed with FeSO4 under CP conditions exhibited higher abundances of *Methanobrevibacter smithii*, *Clostridium* spp., and *Muribaculaceae* spp ($Q < 0.05$). By day 42, pigs fed with FeAA under both sanitary conditions and those fed with FeSO4 under CP conditions showed higher abundances of *Oribacterium* spp. and *Kiritimatiellia* spp. These findings indicate that feeding FeAA under different sanitary conditions may reduce post-weaning diarrhea compared to conventional Fe sources, while also modulating the gut microbiome. **Key Words:** microbiome diarrhea iron

**37 *In vitro* comparison of monovalent copper oxide and traditional copper sources on bacterial control and microbial function.** Y.J.Y. Manaig*[1], W. Vahjen[2], S. Durosoy[1], and A.R. Monteiro[1], [1]*Animine, Annecy, France*, [2]*Freie Universität Berlin, Berlin, Germany*. Copper (Cu) is widely used in animal production for its antimicrobial properties and growth-promoting effects, but different sources vary in bioavailability and antibacterial efficacy. *Streptococcus suis* is a significant pathogen causing severe post-weaning infections in piglets, leading to reduced growth and increased mortality. This study aimed to evaluate the antibacterial activity of various Cu sources on *S. suis* growth. The Cu sources tested included copper sulfate (Manica Cobre S.L., Spain), chelated Cu (ACu, AvailaCu®, Zinpro, USA), tribasic Cu chloride (TBCC, IntelliBond®C, Selko, USA), and monovalent Cu (CR, CoRouge®, Animine, France). The antibacterial activity was assessed using the pathogenic strain *S. suis* (DSM 9682) in Brain-Heart-Infusion medium, with each Cu source saturated at 10g/L at a concentration of 32 µg Cu/mL. Microbial growth was monitored by measuring turbidity every 5 minutes over 24 hours, expressed as optical density at 690 nm. Minimum inhibitory concentrations (MICs)—the lowest concentrations of Cu (in µg/mL) that completely inhibited visible growth—were determined after 24 hours of incubation. Data were analyzed using one-factorial ANOVA with Bonferroni post-hoc tests to identify significant differences among Cu sources. Results showed

that CR exhibited the strongest inhibitory effect against *S. suis*, with MIC observed at significantly lower Cu concentration of 64 µg/mL compared to 256 µg/mL for divalent Cu sources. Growth in *S. suis* was significantly delayed, beginning after 9 hours in CR, while growth in the divalent Cu sources started after approximately 3 hours ($P < 0.05$). These findings align with prior studies highlighting the superior antibacterial properties of monovalent copper compared to divalent copper. In conclusion, the *in vitro* trial demonstrated that the monovalent copper had the highest inhibitory potential against *S. suis*, highlighting the need for further research to evaluate its efficacy *in vivo.* **Key Words:** copper, monovalent, *Streptococcus suis*

**38 Effects of dietary nitrogen on the ileal and fecal microbiome in ileal-canulated pigs.** Q. Wang*[1], Z. Rao[1], J. Remus[1], D. Lopez[2], and C. Paulk[2], [1]*International Flavors and Fragrances Inc., New York, New York, USA*, [2]*Kansas State University, Manhattan, Kansas, USA*. This study examined the influence of dietary nitrogen on the ileal and fecal microbiome of ileal-cannulated pigs, as the microbial profile of pigs fed diets without N sources has not been well researched, and the relationship between basal endogenous N loss and microbial profile is unknown. Seven pigs (241×600, DNA, Columbus, NE; 36.0 to 84.1 kg) were surgically fitted with a T-cannula in the distal ileum. After a 7-d surgical recovery period, the individually housed pigs were allotted to 1 of 2 experimental diets using a Latin square design. The pigs were used for a total of 5 periods of 7 d each, resulting in 20 replicate pigs for the corn-SBM diet (4 replicates per period) and 15 replicate pigs for the N-free diet (3 replicates per period). There was a 7-d washout period between each experimental period. Four types of samples were collected: ileal swab, ileal digesta, fecal swab, and feces at the end of each experimental period. 16S amplicon sequencing was used to identify the microbial communities. PERMANOVA test was used to quantify multivariate community-level differences between treatments. At the genus level, there were significant differences ($P < 0.01$) between pigs fed the N-free diet and those fed the corn-SBM diet for all sample types. Pigs fed the N-free diet showed a distinct microbial profile compared to pigs fed the corn-SBM diet. Differential abundance analysis revealed that 3 genera (*Bacteroides*, *Fusobacterium,* and *Phocaeicola)* were significantly more abundant in N-free diet pigs compared to corn-SBM diet pigs in all sample types ($P < 0.05$). In summary, the microbial profile of pigs fed the N-free diet was significantly different from pigs fed the corn-SBM diet. This suggests that using N-free diets for the estimation of standard basal endogenous N loss in digestibility research may need further evaluation, as the differences of microbial profiles between pigs fed N-free diet and corn-SBM diet may result in different levels of basal endogenous N loss. **Key Words:** digestibility, microbiome, pigs

**39 The effect of thermal treatment of field peas on intestinal microbiota and bacterial metabolites in weaned piglets'.** J. Schulze Holthausen*, W. Vahjen, and J. Zentek, *1Institute of Animal Nutrition, Department of Veterinary Medicine, Freie Universität Berlin, Berlin, Berlin, Germany*. Carbohydrate fractions of legumes may affect the microbiota in the gastrointestinal tract of pigs, resulting in a higher microbial diversity in the large intestine and a more resilient state in the small intestine. The aim of this study was to investigate the effect of thermal treatment on field peas fed to weaned piglets with regards to their effects on the ileal and colonic microbiota and their metabolites. Piglets (n = 48), were weaned with 35 d of life and fed balanced isocaloric and isonitrogenic weaning diets containing 20 % grounded peas (Pea), steamed at 85°C for 10 min (Pea-S), autoclaved at 110°C for 15 min (Pea-A) or a diet with soybean meal (Con). After two weeks 24 male piglets were euthanized and ileal and colonic digesta were snap frozen in liquid nitrogen. DNA was isolated and analyzed through amplicon sequencing targeting 16S rDNA. Statistical analyses were performed using Kruskal-Wallis-Test, followed by Mann – Whitney – U-Test, ($P \leq 0.05$) indicating significant differences ($P \leq 0.05$) and ($P < 0.1$) trends. Microbial diversity indices showed a significantly higher microbial richness in colon digesta of Pea-A compared to Con fed piglets ($P = 0.043$). Shannon-Index was significantly higher in piglets fed peas compared to piglets fed with Con ($P = 0.046$)**.** In colon digesta the relative abundance of *Faecalibacterium* in Pea fed piglets ($P = 0.019$) and the relative abundance of *Ruminoccus 2* in Pea-A fed

piglets ($P$ = 0.003) was significantly higher compared to Con fed piglets. No significant effect of the different feeding groups on bacterial metabolites were visible. Results demonstrate an increase in microbial diversity with the inclusion of peas in the diet of male weaned piglets, which is even more pronounced when peas are thermal treated. Inclusion of peas in the diet of male weaned piglets increased genera which are associated with overall health and growth of pigs, like *Faecalibacterium* and *Ruminococcus 2*. **Key Words:** pea, microbiota, heat treatment

**40 Dietary fiber fermentability and weaning age shape gut microbiota composition and activity in piglets.** L Grzeskowiak, F Ghazisaeedi, M Fulde, J Schulze Holthausen, B Martinez-Vallespin, W Vahjen, and J Zentek*, *Freie Universität Berlin, Berlin, Germany*. Nutritional interventions play a vital role in maintaining the intestinal health of pigs. The fermentability of dietary fiber has the potential to influence gut microbiota diversity and activity in pigs. Weaning age is another critical factor for piglet health. This study hypothesizes that the source of dietary fiber in sow and offspring diets, combined with weaning age, affects gut microbiota composition and activity in piglets. Sows were fed balanced, isocaloric, and isonitrogenous experimental diets that contained 10% of either hay (n = 10) or sugar beet pulp (SBP) (n = 11) during gestation and lactation. Their piglets were weaned at 28 (conventional weaning, CW) and 42 days (late weaning, LW) and received starter diets with fiber sources as their dams. Two weeks later, they were dissected and colon digesta was collected for DNA extraction, 16S-rDNA sequencing and short-chain fatty acid analysis (SCFA) via gas chromatography. Sequences were analyzed using Linear Discriminant Analysis Effect Size (LEfSe). SCFA data were analyzed using SPSS v.29. Statistical significance was at $p \leq 0.05$. The LEfSe analysis revealed *Bulleidia* was more abundant in piglets fed hay, while *Prevotella* predominated in SBP-fed piglets. *Akkermansia*, *Faecousia* and *Blautia* predominated the gut microbiota of CW piglets, while *Clostridium*, *Streptococcus*, *Megasphaera* and *Terrisporobacter* predominated the microbiota of LW piglets. Acetate, n-butyrate and total SCFA levels were higher in LW piglets fed SBP *vs*. hay diets ($P \leq 0.05$). n-Butyrate and n-valerate were higher in LW *vs*. CW piglets fed hay ($P \leq 0.05$). Total SCFA levels including acetate, propionate, n-butyrate and n-valerate were significantly higher in LW *vs*. CW piglets fed SBP ($P \leq 0.05$). Sow and piglet diets high in fermentable fiber and delayed weaning profoundly shape piglet gut microbiota and metabolic activity. These findings highlight the critical role of the sow-offspring association, nutrition and weaning in offspring gut health and development. **Key Words:** hay, sugar beet pulp

**41 Assessing the intestinal diarrhea through *Lactobacillus*, coliforms and *E. coli* population isolated from stool samples.** Tran Thi Quynh Lan*, Do Thien Thai, and Tran Thi Dan, *Faculty of Animal Science and Veterinary Medicine, Nong Lam University- Ho Chi Minh City, Ho Chi Minh city, VietNam*. The purpose of study was to evaluate the intestinal status of piglets by analyzing the number of *Lactobacillus* and *E. coli*. *Lactobacillus:coliforms* ratio (L/C ratio) was applied to assess the risk of developing diarrhea in different stages post-weaning in piglets. This research was carried out at two farms in practice. The stool samples were collected respectively 5 times at 25 days old (piglets weaned at 26 days of age) and at the post-weaning stage (27, 32, 39 and 44 days of age). A total of 100 samples were collected (10 samples/time/ farm). The total *Lactobacillus*, coliforms, and *E. coli* were counted from fecal samples on specific mediums for bacteria culture (MacConkey Agar to coliforms/*E. coli*, MRS agar to *Lactobacillus* in anaerobic condition). Counts were recorded as colony-forming units per gram (cfu/g) and the CFU was changed from cfu/g to log (cfu/g) in statistical analysis. The number of *Lactobacillus* bacteria at 7.80 log CFU/g (farm A) and 7.98 log CFU/g (farm B) tended to decrease at the 1st day post-weaning (pig at 27 days old) at both pig farms ($P < 0.1$). At 6 days post-weaning (pig at 32 days of age), *Lactobacillus* bacteria remained stable between 8.05 log CFU/g and 8.50 log CFU/g until the piglets reached 44 days of age. The number of coliforms showed between 6.01 log (CFU/g) and 6.55 log (CFU/g) throughout the survey period. The number of *E. coli* (approximately 6 log CFU/g) was below the pathogenic *E. coli* (F4 or F18) number could cause diarrhea in the piglets (7 log CFU/g). In the pre-weaning period, 20% of the fecal samples showed the L/C ratio ≤ 1.3 in both farms, indicating that the risk of diarrhea was

low, however 60% of fecal samples showed this proportion was lower than 1.3 at one day post-weaning. This indicated a risk of diarrhea in the weaning piglets. Therefore, the use of nutritional interventions targeted at the gut microbiome should be considered to rebalance the commensal microbiome in the gut of weaning piglets. **Key Words:** L/C ratio, *E. coli*, weaning

**42 Effects of Combining Garlic with Apple Pomace or Blackcurrant on the Gastrointestinal Microbiome of Organic Pigs After Weaning.** K Jerez-Bogota*[2,1], M Jensen[1], O Højberg[2], and N Canibe[2], [1]*Department of Food Science, Aarhus University, Aarhus, Denmark*, [2]*Department of Animal Science, Aarhus University, Tjele, Denmark*. We have previously shown that combinations of antibacterial plants alleviate postweaning diarrhea caused by enterotoxigenic *Escherichia coli* (ETEC). Here, we evaluated the effect of feeding garlic combined with either apple pomace or blackcurrant on the gastrointestinal ecosystem of organic weaners three weeks after an ETEC challenge at weaning. The study involved two blocks of 32 pigs (7 wk old), housed in pairs. Pens were randomly assigned to four treatments: non-challenged (NC), ETEC-challenged (d1-2; $10^9$ CFU/ml ETEC-F18 9910297-2$^{ST}$™; PC), ETEC-challenged fed garlic and apple pomace (3% w/w each; GA), and ETEC-challenged fed garlic and blackcurrant (3% w/w each; GB). One pig per pen was euthanized on days 21–22 after weaning for digesta and mucosa sampling. Digesta was analyzed for bacterial counts (qPCR), microbial profiling (16S rRNA), and metabolites, while jejunal and ileal mucosa were assessed for antioxidant enzyme activity. Compared to NC, PC had a greater abundance of *Campylobacter* and lower of *Roseburia* in cecum ($P < 0.05$). GA had greater abundance of *Weissella* in stomach and jejunum, *Catenibacterium* and *Faecalibacterium* in cecum, and *Holdemanella* in colon, compared to NC ($p < 0.05$). Further, compared to NC, GA had lower lactic acid along the gut, lower cadaverine in the stomach and ileum ($P < 0.05$), and higher butyric acid in colon ($P < 0.05$). Similarly, GB had a greater abundance of *Catenibacterium* and *Holdemanella*, along with higher total short-chain fatty acid levels in the cecum and colon and higher butyric acid in the colon ($P < 0.05$), compared to NC. Superoxide dismutase and glutathione peroxidase activities in jejunal and ileal mucosa were higher in GA and GB than in NC and PC ($P < 0.05$). In conclusion, the combination of garlic with either apple pomace or blackcurrant increased the abundance of beneficial gut bacteria, leading to improved hindgut fermentation activity and oxidative status in the small intestine of pigs three weeks after weaning. **Key Words:** antibacterial plants, microbiome, postweaning

**43 Novel pathway activation mapping to characterize the physiological effects of *Salmonella* infection in piglets.** M. C. Walsh*, L. Payling, and L. F. Romero, *Biofractal, Loule, Portugal*. The objective was to reanalyse blood and ileum gene expression data from previous trials (Huang et al. 2018; KyeongHye et al. 2022) with novel methods that quantified changes in biological pathways associated with *S. Typhimurium* challenge in piglets. The dataset included 6 ileal and 8 blood samples from piglets either challenged with *S. Typhimurium* or unchallenged controls. 4 weeks old piglets were orally challenged with $10^9$ CFU *S. Typhimurium*. Gene expression data determined via RNA sequencing were aligned to the *Sus scrofa* reference genome (Sscrofa11.1) using Kallisto. Data were normalised and tested for differentially expressed genes, followed by a novel topology-based Quantitative Pathway Activation (QPA) method (Biofractal, Portugal) to quantify the activation or inhibition of pathways using a customization of the Reactome database. In the ileum of pigs infected with *Salmonella* there was evidence of activated immune function and compromised gut integrity, especially cell turnover, indicating poor cellular health. Nutrient digestion, absorption and metabolism were inhibited including vitamins, minerals, fats and proteins ($P_{adj} < 0.05$). In the blood of *Salmonella* challenged piglets, immune activation was also detected. However, adaptive immunity was inhibited. Oxidative and cellular stress were activated, as well as cellular apoptosis and senescence indicating poor cellular health. There was strong activation of glycogen metabolism and lipid metabolism. Cell replication and red blood cell function were among the top inhibited functions ($P_{adj} <0.05$). These findings demonstrate that *Salmonella* challenge was characterised by poor gut integrity and reduced nutrient digestion and absorption even 28d after challenge. The

systemic effects of this gut health challenge were clearly visible in blood 7d after challenge. Blood gene expression presents an interesting opportunity to detect and quantify gut health challenges in piglets. **Key Words:** Piglets, Salmonella, Transcriptomics

**44 Dietary supplementation of multi-strain probiotics alters the fecal microbial profile and improves the carcass quality in commercial pigs.** Jai-Wei Lee*[1], Ting-Yu Lee[2], Shi-Yong Liu[1], and Jin-Seng Lin[2], [1]*Department of Tropical Agriculture and International Cooperation, National Pingtung University of Science and Technology, Neipu, Pingtung, Taiwan*, [2]*SYNBIO TECH INC, Kaohsiung, Taiwan*. The objective of the present study was to investigate effects of supplementing multi-strain probiotics, including *Lactiplantibacillus plantarum*, *Streptococcus thermophilus*, and *Bacillus*, in feed on the growth performance, fecal microbial profile, and carcass quality of commercial crossbred pigs. A total of 144 weaning piglets were randomly assigned to two treatment groups (18 piglets /pen, 4 pens/group) and fed basal diet with (Pro) or without (Control) dietary supplementation of multi-strain probiotics ($1\times10^6$ CFU/g feed), respectively, for 18 weeks until reaching the market weight (~115 kg). At the end of feeding trial, 6 pigs were randomly selected from each group and subjected to analysis of fecal microbial profile and carcass quality. Results indicated that parameters related to growth performance, including body weight gain and feed conversion rate, were not different ($P > 0.05$) between two groups. In terms of carcass quality, pigs from Pro group had significantly ($P < 0.05$) increased arm weight (10.74 vs. 9.74 kg), blade shoulder weight (5.66 vs. 4.97 kg), and larger loin eye area (65.18 vs. 53.20 $cm^2$) when compared to those from the control group without compromising parameters related to meat quality ($P > 0.05$). Analysis of 16S rRNA gene sequencing on fecal microbiota composition and bacterial abundance demonstrated that the amounts of butyrate-producing bacteria *Atopobiaceae* ($P = 0.089$) and *Veillonellaceae* ($P < 0.05$) were more enriched in pigs from Pro group. Moreover, results from examining the expression of genes involved in insulin and mTOR signaling pathways in loin eye samples indicated that supplementation of probiotics notably upregulated protein synthesis, leading to increased lean muscle growth. In conclusion, dietary supplementation of multi-strain probiotics could improve certain carcass traits without compromising growth performance and meat quality. **Key Words:** Carcass quality, Probiotics, Butyrate

**45 Effect of fermented cereal liquid feed supplemented with *Pediococcus acidilactici* on gut microbiota, mucosal immunity, and growth in suckling and post-weaning piglets.** J. Xu*[1], A.A Schönherz[1], K.S. Jerez-Bogota[1], S.J. Noel[1], K. Skovgaard[2], P.M.H. Heegaard[3], C. Lauridsen[1], H.N. Lærke[1], and N. Canibe[1], [1]*Department of Animal and Veterinary Sciences, Aarhus University, Tjele, Denmark*, [2]*Department of Biotechnology and Biomedicine, Technical University of Denmark, Lyngby, Denmark*, [3]*Department of Health Technology, Technical University of Denmark, Lyngby, Denmark*. This study investigated the effects of fermented cereal liquid feed supplemented with *Pediococcus acidilactici* (FCLFp), introduced in suckling period, on gut microbial ecology and gut health pre- and post-weaning. The strain was selected based on our in vitro study. Ninety piglets were divided into two groups 1) DRY: fed with standard dry feed (n = 42), 2) FERM: fed with FCLFp (n = 48). Piglets had *ad libitum* access to their diets from d14 to d56 and were weaned on d28. Fecal and blood samples (FERM n = 24, DRY n = 21) were collected weekly, starting d14, for diarrhea evaluation, microbiota and metabolites analysis; digesta and gut mucosa (FERM n = 8, DRY n = 7) were collected on d7 post weaning (pw), for microbiota composition (16S rRNA sequencing), metabolites, and gene expression analysis. FERM had higher average daily feed intake (ADFI) ($P < 0.05$) the first week pw. On d7 pw, FERM showed increased expression of transcripts coding for glucose transporter 2, a tendency for increased pro- and anti-inflammatory cytokines expression (IL10, IL8, IL17; $P < 0.10$), and reduced mucin 2 expression in the jejunal mucosa, along with higher gastric lactic acid and acetic acid levels, and lower pH. The two groups had different microbiota compositions (Bray-Curtis) in the stomach, cecum and colon. Two weeks pw, FERM showed greater soft stools occurrence ($P < 0.05$) indicating diarrhea, decreased fecal microbiota diversity (Shannon index) and increased fecal biogenic

amine concentration (cadaverine and agmatine, $P < 0.05$), and a tendency to have higher levels of fecal *E. coli* and lower ADFI ($P < 0.10$), all suggesting dysbiosis. Four weeks pw, FERM recovered from diarrhea and moved towards a balanced gut microbiota (similar α-diversity and composition in both groups), with a higher average daily gain and ADFI than DRY ($P < 0.05$). In conclusion, although FCLFp seemed to negatively affect indicators of gut health the first two weeks pw, it supported growth performance four weeks after weaning. **Key Words:** Fermented liquid feed, post-weaning diarrhea, gut microbiota

**46 Particle size of cereals shapes piglet gut microbiota during *in vitro* fermentation.** V. H. Tran*[1], X. Liu[2], B. M. Flanagan[2], B. A. Williams[2], G. Feng[1], M. Navarro[1,2], E. Roura[1,2], M. J. Gidley[2], X. Wu[2], L. Omaleki[1], and C. Turni[1], [1]*Centre for Animal Science, Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, St Lucia, Queensland, Australia*, [2]*Centre for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, St Lucia, Queensland, Australia*. Optimising ingredients, particle sizes and the use of exogenous enzymes in diet to improve post-weaning piglet's digestibility is a common practice. However, their effects on gut microbiota remain unexplored. This study investigated the fermentation traits of combinations made from different ingredients, particle sizes, and enzyme treatments using *in vitro* batch fermentation with piglet faecal inoculum. Sixty samples were prepared from six ingredients including cereals (barley, maize, wheat) and protein sources (soybean meal, canola meal, lupin), two particle sizes (large 1.18 – 2.36 mm and small 0.212 – 0.425 mm), and five enzyme treatments (no enzyme - control, phytase, protease, xylanase and glucanase, and a mixture of all enzymes). Samples were digested *in vitro* following the INFOGEST protocol, washed using 0.1 mm filter, and freeze-dried. Starch content of digesta was analysed before anaerobically fermenting *in vitro* for 48 hours at 39°C with basal solution, bicarbonate buffer, reducing agent, vitamin/phosphate solution and inoculum prepared from faeces of pre-weaning piglets. Microbial profiling was performed using 16S rRNA sequencing. Large-particle cereals retained threefold more starch post-digestion than small-particle cereals and protein sources ($P < 0.05$). This promoted the dominance of *Succinivibrio*, a starch degrading genus, reducing the microbial diversity (twofold lower Shannon and Simpson indices, $P < 0.05$). Conversely, hemicellulose-degrading genera such as *Fibrobacter*, *Prevotella_9* and *Bacteroides* were prevalent in small-particle cereals and protein sources. In conclusion, large-particle cereals stimulated starch fermentation, while small-particle cereals and protein sources favoured insoluble fibre fermentation. The particle size impact on gut fermentation highlights the need to consider particle sizes in piglet diet formulation to promote healthy gut microbiota. **Key Words:** *in vitro* fermentation, gut microbiota, feeds utilisation

## Theme III Posters: Functional Ingredients and Utilization of Feed Resources for Improved Digestive Function and Nutrient Efficiency

**47 Threonine, tryptophan and valine as functional amino acids for improving growth performance of piglets during nursery in a natural disease challenge model.** M.V. Curtasu*[1,2], B. Yanibada[2], A.R. Alfonso Avila[3], A. Simongiovanni[4,5], T Chalvon-Demersay[5], and M.P. Létourneau-Montminy[2], [1]*Aarhus University Viborg, Department of Animal and Veterinary Sciences, Tjele, 8830, Denmark,* [2]*Laval University, Faculty of Agriculture and Food Sciences, Department of Animal Sciences, 2425 rue de l'Agriculture, Québec, G1V 0A6, Canada,* [3]*Deschambault Animal Science Research Centre (CRSAD), 120-A chemin du Roy, Deschambault, G0A 1S0, Québec, Canada,* [4]*METEX ANIMAL NUTRITION, Paris, France,* [5]*EUROLYSINE, Paris, France*. The objective of this study was to evaluate the effects of amino acids as functional ingredients in weaning pigs under a natural disease challenge with porcine reproductive and respiratory syndrome virus (PRRSv). A control diet (CON) was compared to a functionally enriched amino acid diet (AA+), which included a 10% increase in all essential amino acids, with additional increases in threonine (+15%), tryptophan (+40%), and valine (+20%). A total of 300 piglets (five consecutive batches of 60 piglets at 21 days old, PRRS-positive, from different sows) were assigned to either CON (n = 150) or AA+ (n = 150) diets *ad libitum* for 4 weeks. Body weight was measured daily during the nursery phase and health status was assessed every week from day 0 to day 28. Untargeted metabolomics was used to investigate the plasma, and a subset of piglets was selected (n=60) for sequencing of the fecal microbiota. Zootechnical parameters were analyzed using a linear mixed-effects model in SAS. Metabolomics and microbiome were analyzed using multivariate data analysis. The AA+ diet resulted in higher body weight (+5.3%, $P < 0.01$), average daily gain (+12.2%, $P < 0.01$), and average daily feed intake (+7.4%, $P < 0.01$) compared to the CON. A decline in health scores occurred from day 0 to day 14 ($P < 0.001$), regardless of the diet. Plasma metabolomics showed an enrichment in amino acids particularly threonine and valine, and increased levels of tryptophan metabolites (indole-3-carboxaldehyde, 3-indoxyl-sulphate) with AA+ diet. Fecal beta-diversity was modulated (PERMANOVA, $P = 0.046$) and based on a Linear Effect Size Analysis (LefSe) members of the genus *Blautia, Subdoligranulum, Coprococcus, Prevotella* and *Fournierella* were significantly higher in the AA+ compared to CON ($P < 0.05$, FDR-adjusted). In conclusion, a functionally optimized amino acid-enriched diet contributed to an improved growth performance in PRRS-positive pigs with visible effects in the plasma metabolome profile and gut microbiome. **Key Words:** amino acids, metabolomics, microbiota

**48 Supplementation of a consensus bacterial 6-phytase variant on reproductive performance of sows fed diets without added inorganic phosphate and reduced energy and nutrients over two cycles.** Deepak E. Velayudhan*[1], Georg Dusel[2], Ester Vinyeta[1], Leon Marchal[1], and Yueming Dersjant-Li[1], [1]*Danisco Animal Nutrition & Health (IFF), Oegstgeest, The Netherlands,* [2]*University of Applied Sciences Bingen, Bingen am Rhein, Germany*. The study evaluated if a consensus bacterial 6-phytase variant (PhyG) could maintain reproductive performance in sows fed no added inorganic phosphate (iP), energy and nutrient-reduced diet through 2 full, sequential reproductive cycles from service of the first cycle to weaning of the second cycle. Ninety sows with an average parity of 3.1 and average initial body weight (BW) at insemination of 211.1±11.1 kg, were randomly assigned, based on parity, over 2 experimental diets soon after confirming pregnancy. Treatments included an energy and nutrient-adequate positive control (PC) diet, a negative control (NC) diet without added iP, reduced in Ca, net energy, digestible amino acids and Na (based on the contribution of 1000 FTU/kg phytase), supplemented with PhyG at 1,000 FTU/kg. Diets were based on barley, wheat, corn, soybean meal and sunflower meal (phytate P of 2.8 g/kg) with restricted feeding during gestation and ad libitum during lactation. The reproductive performance of sows and the growth performance of their litters were recorded. Data were analyzed using ANOVA, using JMP 16.1 with cycle as a random effect. Phytase supplementation to the energy and nutrient-reduced, iP-free diet did not affect ($P > 0.10$) the sow BW gain during gestation,

BW loss from farrowing till weaning, sow body condition score and the backfat thickness measured at different time points compared to PC. Supplementation of PhyG did not affect the piglet response parameters ($P > 0.10$), including the number of piglets born, piglets born alive, stillborn, mummified piglets, piglet birth weight, and piglet mortality. Moreover, PhyG supplementation showed a higher ($P < 0.05$) BW gain of piglets at weaning vs PC (329 vs 279 g). In conclusion, PhyG supplementation to an iP-free, energy and nutrient-reduced diet maintained reproductive performance in sows and litter performance, along with a 6.5% reduction in the feed cost due to the matrix application. **Key Words:** consensus bacterial 6-phytase variant, reproductive performance, sows

**49 Impact of inulin supplementation and animal density modulation on intestinal health parameters in weaned piglets.** P. Siegien*[1], M. Habets[1], M. Gillis[1], J. Wavreille[2], J. Bincelle[1], and M. Schroyen[1], [1]*Gembloux Agro-Bio Tech, Precision Livestock and Nutrition Laboratory, TERRA Teaching and Research Centre, Gembloux Agro-Bio Tech, University of Liège, 5030 Gembloux, Belgium*, [2]*Walloon Agricultural Research Centre, Animal production unit, Walloon Agricultural Research Centre, 5030 Gembloux, Belgium*. Social stress in piglets poses welfare and production issues for the pig industry. Through the gut-brain axis, social stress relates with intestinal health. This study evaluated the combined effects of inulin and chronic social stress on molecular, physiological, and production-level indicators of intestinal health in piglets. A total of 144 piglets, sex ratio 50:50, were weaned at 4 weeks of age and fed for 4 weeks a control or inulin diet (nutrient adequate with 0 or 10g inulin/kg feed) in 12 pens with high (n = 14) or low (n=10) animal densities (2.75 vs 1.96 pigs/m²). At 8 weeks of age, 12 piglets/treatment were slaughtered for intestinal sampling. Inflammatory and permeability gene expression (qPCR) and histomorphological structure were analysed in intestinal tissue. Short-chain fatty acids (SCFA) were measured (HPLC) in intestinal content. Outliers were removed using QQPlot, and ANOVA was performed. Body weight, feed intake and histomorphology showed no differences between treatments. In the colon, *BAX*, *IL1α* and *IL6* expression decreased with inulin ($P < 0.05$) but *IL1α* increased in high density pens ($P < 0.05$). No diet-density interaction was found for any of the parameters. In the ileum, *TANK* expression decreased with high density ($P < 0.05$). SCFA profiles showed higher molar ratio of isobutyric acid in the colon and higher acetic acid molar amounts in the jejunum of inulin fed pigs while high density lowered molar amounts of propionic acid in the jejunum ($P < 0.05$). Inulin showed anti-inflammatory effects while, oppositely, high animal density activated the NFκB pathway. Despite molecular signs of social stress and the effect of inulin, piglet performance was minimally impacted, likely due to insufficient stressor and a low inulin inclusion rate. No significant interactions between inulin and density were found, suggesting each affects intestinal health independently. Addressing high density stress remains critical for improving gut health and performance in piglets. **Key Words:** Animal density, Gut-brain axis, Social stress

**50 Mycotoxin mitigation strategies against the emerging mycotoxins enniatins on suckling and nursery piglet performance.** S. van Kuijk*[1], G. Wang[1], A. Middelkoop[2], R.R. Santos[2], and H.V.L.N. Swamy[1], [1]*Trouw Nutrition, Stationsstraat 77, 3811 MH Amersfoort, The Netherlands*, [2]*Schothorst Feed Research, Meerkoetenweg 26, Lelystad, The Netherlands*. The study aimed to test two mycotoxin mitigation strategies against emerging mycotoxins, enniatins (ENN), fed to piglets from birth up to 35 days post-weaning. Litters of 4 sows per treatment were enrolled from birth onwards. Mycotoxin levels in the sow diets were negligible and ENN were absent in the diets. Three feeding phases included creep feed (d0-28 of age), post-weaning phase 1 (d0-14) and phase 2 (d14-35). From d3 of age up to end of the trial, piglets were fed one of the four experimental diets: 1) control diet (CON), 2) control diet with naturally contaminated barley aiming at 0.8 ppm ENN in the final diet (ENN), 3) ENN diet with 1 kg/t TOXO-XL (Selko, Tilburg, The Netherlands) (ENN+TP1), 4) ENN diet with 1 kg/t TOXO-Innovation (ENN+TP2). After weaning, at d28, piglets were mixed within pre-weaning treatment and housed in mixed sex groups of 5 animals/pen and studied up to d35 post-weaning. Body weight (BW) and feed intake were measured on

d3, at weaning, and weekly post-weaning. Data was analyzed using ANOVA and treatments were compared using Tukey post-hoc in Genstat. Low mortality (2 pigs), low medication use (5 pigs) and no diarrhea were observed throughout the study. BW at weaning was ($P$ = 0.03) lower in the ENN treatment (7.1 kg) compared to the CON treatment (7.9 kg) with the other two treatments being intermediate, similarly for pre-weaning ADG ($P$ < 0.01). D0-7 post-weaning ADG was lowest ($P < 0.01$) for ENN+TP1 followed by ENN and ENN+TP1 and CON. This difference started waning reaching same ADG (404 g/d) for ENN and CON by d35 post-weaning. ENN+TP1 (451 ± g/d) and ENN+TP2 (470 g/d) had varying positive response over time but overall performance of both treatments was significantly better ($P$ < 0.001) than both CON and ENN. Overall FCR was higher ($P$ < 0.001) for CON (1.39) compared to the other treatments (1.28-1.32). In conclusion, mycotoxin mitigation products can improve piglet performance when fed diets with (marginal) ENN contamination. **Key Words:** Mycotoxins, Enniatins, Growth performance

**51 The impact of protein fermentation on intestinal health in pigs.** L Noorman[1], S de Vries*[2], and WJJ Gerrits[2], [1]*Department of Population Health Sciences, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands*, [2]*Animal Nutrition Group, Wageningen University & Research, Wageningen, The Netherlands*. *In vitro* and *in situ* experiments have demonstrated that metabolites produced during protein fermentation can negatively affect intestinal health. However, *in vivo* research on their impact on intestinal health remains limited. This study aimed to investigate the effects of protein-derived metabolites on fecal consistency and intestinal permeability in pigs. A total of 128 boars were fed a diet containing bovine collagen (BC), zein (ZE) as low-digestible-, or whey protein isolate (WPI) as highly-digestible protein source, and addition of no- (NFF), rapidly- (RFF), or slowly fermentable fibers (SFF). Fecal consistency was monitored daily, and after two weeks, colon digesta samples were analyzed for titanium, ammonia ($NH_3$), volatile fatty acids (VFA), biogenic amines, and indolic and phenolic compounds. Jejunal and colonic segments were collected to assess intestinal permeability to 4 kDa-FITC and 40 kDa-TRITC dextran. Data were analyzed using two-way ANOVA with protein, fiber, and batch as factors, principal component analysis, and Pearson correlation coefficients. Fecal consistency scores declined over time for BC but remained relatively constant for WPI resulting in lower scores for BC from day 7 onwards (29-46%, $P$ < 0.05), regardless of the presence of fibers (protein × fiber, $P$ > 0.05). Less consistent differences were observed for ZE. No correlations were found between protein-derived metabolites and fecal consistency or intestinal barrier function, except for ammonia. Colonic ammonia flow was positively correlated with colonic permeability to 4 kDa-FITC dextran (proximal colon: $r$ = 0.65, $P$ < 0.001; distal colon: $r$ = 0.59, $P$ < 0.001) and 40 kDa-TRITC dextran (proximal colon: $r$ = 0.78, $P$ < 0.001, distal colon: $r$ = 0.71, $P$ < 0.001). In conclusion, while low-digestible proteins reduced fecal consistency, no causal link was found with protein-derived metabolites. Among all metabolites, only ammonia appeared to impair intestinal barrier function. **Key Words:** protein-derived metabolites, intestinal permeability, fecal consistency

**52 In vitro gastric and intestinal protein digestion kinetics in high-protein sunflower meal or soybean meal-based diets without or with exogenous phytase.** F Njeri*[1], M Anh Ton Nu[2], H Schulze[3], and E. G Kiarie[1], [1]*University of Guelph, Guelph, Ontario, Canada*, [2]*AB Neo, Videbaek, Denmark*, [3]*Livalta, Peterborough, UK*. Soybean meal (SBM) and high-protein sunflower meal (SFM) had shown comparable crude protein (CP) solubility, but both are high in phytate, which may limit CP digestibility. This study aimed to evaluate the effect of protein source and exogenous phytase addition on *in vitro* CP digestion kinetics of SBM- and SFM-based diets. Cornstarch-based diets were formulated to contain 40% of either SBM or SFM with 0 or 500 FTU/kg phytase. Sample (500 mg CP, triptricate) were incubated with porcine pepsin at pH 3.5 and 39°C for 0, 0.5, 1, and 1.5h in gastric phase, followed by small intestine digestion with porcine pancreatin and bile extract at pH 6.8 for 0, 0.5, 2, 4, and 6h. In vitro protein digestion kinetics were modeled using a Gompertz function in SAS. Data were analyzed with protein source and phytase as fixed effects and their interaction. Dietary CP was classified based on their digestion time as

$CP_{fast}$ (0-0.5h), $CP_{slow}$ (0.5-4h), and $CP_{resistant}$ (>4h). SBM and SFM had 51.4 and 52.4% CP, 0.48 and 0.54% phytate P, 17.0 and 16.6% insoluble fiber, 9.4 and 5.5% soluble fiber on DM, respectively. Digested CP in SFM diet was 2.2-4.4 times higher in stomach and 1.1-3.2 times higher in small intestine (except 0.5 and 6h) than SBM diet depending on digestion time ($P < 0.05$). Phytase addition increased soluble CP of SFM at 1h in stomach and of SBM at both 0.5h and 6h in small intestine ($P < 0.05$). The interaction ($P < 0.05$) between phytase and protein source in small intestine was such that, phytase addition increased $CP_{fast}$ by 28% and reduced $CP_{slow}$ by 75% in SBM but only decreased $CP_{resistant}$ by 25% in SFM ($P < 0.05$). SFM also showed 16% more total digested CP (A) than SBM. Phytase addition increased digestion rate ($K_u$) and inflection digestion time ($T_i$) in SBM but not in SFM. In conclusion, SFM have digestible CP compared to SBM and phytase addition further increased in vitro protein digestion in SBM due to processing. Further in *vivo* amino acids digestibility study is recommended. **Key Words:** Processed sunflower meal, in vitro digestibility, Digestion kinetics

**53 Effects of a combination of protease and multi-strain *Bacillus* spp. direct fed microbial supplementation on the growth performance of weaned pigs fed a high fiber diet.** P. Aymerich*[1], D. E. Velayudhan[1], M. Rodríguez[2], L. Marchal[1], and E. Vinyeta[1], [1]*Danisco Animal Nutrition & Health (IFF), Oegstgeest, The Netherlands*, [2]*Animal Data Analytics, S.L., Segovia, Spain*. The hypothesis of this study was that fiber degrading enzymes produced by certain spore-forming *Bacillus* spp. can hydrolyze dietary fiber when spores germinate in the small intestine. These enzymes in combination with a protease could increase energy and nutrient digestibility, resulting in improved growth performance in pigs. Therefore, this study evaluated a combination of protease and Bacillus spp. multi-strain direct-fed microbial on the growth performance in weaned pigs fed a high fiber diet. A total of 256 weaning piglets ([Pietrain × Danbred], 28 days of age, initial body weight (BW) of 8.4 ±1.1 kg) were used in a randomized complete block design with 2 dietary treatments, with 16 pen replicates/treatment, and 8 piglets per pen (1:1 male: female). Diets were based on corn, wheat, barley, wheat middlings, rapeseed meal and soybean meal and fed in mash form in two phases: pre-starter phase (day 1-14) and starter phase (day 15-42), with a calculated NDF content of 14.3 and 14.7%, respectively. Treatments comprised: 1) control nutritionally adequate diet (CTRL); 2) CTRL supplemented with a protease and multi-strain direct-fed microbial (PRO+DFM), containing protease at 5,000 U/kg and 3-strain of *Bacillus* spp. each at 1.5×10^8 CFU/kg feed. Piglets were individually weighed, and data was used to calculate BW and average daily gain (ADG) per pen. Average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated per pen and corrected for mortality. A one-way ANOVA was performed using JMP (version 16.1). The supplementation of PRO+DFM did not affect final BW (26.5 kg in both treatments) and ADG but tended to reduce ADFI (0.637 PRO+DFM vs. 0.680 kg CTRL; $P < 0.10$). Consequently, FCR improved ($P < 0.05$) during day 14-42 and the overall study period (1.485 PRO+DFM vs. 1.586 in CTRL). In conclusion, a combination of a protease and a multi-strain *Bacillus* spp. direct-fed microbial improved feed efficiency in weaned piglets when fed a high fiber diet. **Key Words:** Protease, Direct fed microbial, Piglet

**54 An integrated analysis to investigate the effects of dietary isoacids supplementation on digestibility, fermentation products, microbiome, and gut pH of ileal-cannulated pigs.** A. F. Bolivar-Sierra*[1], A. P. Benavides-Infante[1], M. T. Socha[2], L. A. Amundson[2], L. Alves Rodrigues[2], B. St-Pierre[1], C. L. Levesque[1], and J. Y. Perez-Palencia[1], [1]*South Dakota State University, Brookings, SD, USA*, [2]*Zinpro Corporation, Eden Prairie, MN, USA*. It has been shown that isoacids supplementation enhanced amino acid (AA) and fiber digestibility, particularly at the ileal level, potentially via microbiome modulation. The objective of this study was to investigate the effects of dietary isoacids supplementation on digestibility, fermentation products, microbiome, and gut pH of ileal-cannulated pigs, using an integrated data analysis approach of two studies. In study 1, pigs were fed normal protein-low fiber diets with or without isoacids (0.0 vs. 0.5–1.5%). In study 2, diets with or without isoacids (0.0 vs. 1.0%) varied in protein and fiber content. Apparent ileal (AID) and total tract digestibility (ATTD) were assessed in both studies, while microbiota and gut pH were analyzed in study 2. Data underwent principal

component analysis (PCA), hierarchical clustering, and partial least squares discriminant analysis (PLS-DA), with features integrated using an adapted DIABLO analysis and visualized with correlation circos plot. Isoacids primarily improved AID of isoleucine (79.2 vs. 81.6±0.02%) and leucine (79.3 vs. 82.1±0.03%) and increased ammonia concentration (253 vs. 287±10ppm), but reduced AID of total dispensable AA (69.3 vs. 67.1±0.02%; $P < 0.05$). Isoacids also improved ATTD of total dispensable (78.2 vs. 80.4±0.02%) and indispensable AA (79.9 vs. 83.3±0.05%; $P < 0.05$). Integration analysis using circos plot revealed that ileal isobutyric, caproic, and isovaleric acids, and fecal valeric, isovaleric, acetic, and butyric acids were modulated by isoacids. A candidate strain, *Romboutsia timonensis*, was identified as a potential modulator of gut responses. Results of the present study suggest that isoacids exert distinct effects at the ileal and total tract levels, likely mediated by specific bacterial species and fermentation products. **Key Words:** digestibility, isoacids, swine

**55 Effects of organic acid-preserved grain on sow and progeny performance, nutrient digestibility, and gut microbiome dynamics from gestation to slaughter.** Shane Maher*[1], Torres Sweeney[2], Stafford Vigors[1], and John V. O'Doherty[1], [1]*School of Agriculture and Food Science, University College Dublin, Ireland.*, [2]*School of Veterinary Medicine, University College Dublin, Ireland.* This study investigated the maternal and direct effects of organic acid (OA)-preserved grain, compared to dried grain, on pig lifetime performance. Eighty sows were assigned to one of two diets based on parity, body weight, and backfat thickness from day 100 of gestation. On day 10 postpartum, their litters were assigned to one of two diets (n=20) in a 2 × 2 factorial: 1) dried grain for both sows and progeny (DD); 2) dried grain for sows and preserved grain for progeny (DP); 3) preserved grain for sows and dried grain for progeny (PD); and 4) preserved grain for both (PP). Pigs remained in these groups post-weaning (PW) and were monitored for 142 days PW. Cereals were either dried to 140 g/kg moisture or preserved with OA mould inhibitor (650 g/kg propionic acid) at storage. Faecal samples were collected at weaning, day 30 PW, and slaughter for total tract digestibility (CATTD) of nutrients and microbial analysis. Pigs from sows offered the preserved grain diet had improved feed conversion ratio (FCR) between days 62-142 PW ($P < 0.05$). Pigs offered preserved grain directly had enhanced daily gain from weaning until slaughter, improved FCR from day 14 PW, and higher BW from day 30 PW onwards ($P < 0.05$). These pigs also exhibited increased CATTD of N and GE ($P < 0.05$). Pigs in the PP group harboured higher diversity at weaning and day 30 PW compared to DD pigs, with increased abundance of beneficial bacteria such as *Ruminococcus* at weaning, *Propionibacterium* on day 30 PW and *Faecalibacterium* at slaughter, compared to DD pigs ($P < 0.05$). In conclusion, offering sows preserved grain during late gestation and lactation enhanced progeny FCR during the finisher stage. Direct feeding of preserved grain from day 10 postpartum to slaughter improved ADG, FCR, and BW, likely driven by enhanced CATTD of nutrients and beneficial microbiome changes throughout production. This demonstrates the potential of OA-preserved grain to improve sustainability and efficiency in pig production. **Key Words:** Organic acids, Cereal preservation, Digestive function

**56 Impact of Superdosed Microbial Phytase on the Fate of Phosphorus and Calcium in Gastrointestinal Tract of Growing Pigs.** J. Labarre*[1,2], A Narcy[3], M. Jlali[4], D.B Bueno Dalto[5], T.D. Crenshaw[6], P. Schlegel[7], and M.P. Létourneau-Montminy[1], [1]*Laval University, Department of Animal Science, Quebec G1V 4G2, Canada*, [2]*Université Paris-Saclay, INRAE, AgroParisTech, UMR MoSAR, 91120, Palaiseau, France*, [3]*INRAE, Université de Tours, BOA, Nouzilly, 37380, France*, [4]*Adisseo France S.A.S, Department of R&I in Monogastric Animal Nutrition, European Laboratory of Innovation, Science and Expertise, 69190 Saint-Fons, France*, [5]*Agriculture and Agri-Food Canada, Sherbrooke R&D Centre, Quebec J1M 0C8, Canada*, [6]*University of Wisconsin, Department of Animal and Dairy Sciences, Madison 53706, Wisconsin, USA*, [7]*Agroscope, Swine Research Unit, Posieux, 1725, Switzerland*. As a part of phytic phosphorus (PP) is not digested with usual dietary inclusion range (500 FTU/kg) of microbial phytase (PhytM), greater concentrations of phytase (superdose), should further improve PP digestion in pigs. Effects of a superdose of PhytM on gastrointestinal tract pH, PP degradation, P and Ca

concentrations and digestibility among the different gastrointestinal tract (GIT) segments were assessed. Twenty-four pigs of 41 kg were randomly assigned to one of the three experimental diets containing 0, 500 or 2000 FTU/kg of PhytM. Control diet without PhytM was formulated to contain 0.34% digestible P and 0.78% total Ca and PhytM, assumed to supply 0.12% digestible P and 0.12% total Ca to both 500 and 2000 FTU/kg diets. After 21 days, pigs were euthanized 2.5h after their last meal and digesta samples from GIT segments were collected. The total P, Ca, PP, and titanium dioxide contents were analyzed in total and soluble fractions and digestibility (disappearance) was calculated. Statistical analysis was done in R using linear (Lin) and quadratic (Quad) contrasts. Dietary differences in pH of the stomach (3.25) and duodenal (5.3) digesta were not detected. The PP degradation was enhanced by PhytM in the stomach (Lin and Quad, $P < 0.001$) and a total of 10, 42 and 62 % of the PP was degraded in the jejunum for diets containing 0, 500 or 2000 FTU/kg respectively (Lin and Quad, $P < 0.001$). In the jejunum, P (Lin and Quad, $P < 0.01$), PP (Lin and Quad, $P < 0.001$), and Ca (Lin, $P < 0.05$) concentrations decreased with the addition of PhytM. In the ileum, PhytM decreased only the P concentrations (Lin and Quad, $P < 0.01$). In the colon, P (Lin and Quad, $P < 0.001$) and Ca (Lin and Quad, $P = 0.06$) digesta concentration were decreased by PhytM. Throughout the colon, P digestibility was improved by PhytM (Lin and Quad, $P < 0.001$) whereas Ca digestibility was not affected. In conclusion, a superdose (2000 FTU/kg) of PhytM improved PP degradation by 1.5 times compared to 500 FTU/kg addition. **Key Words:** phytase, phytic phosphorus, phosphorus

**57 Evaluation of a monoglyceride blend as a sustainable alternative to zinc oxide on nursery pig performance and intestinal health.** A.L.B. Mezzina[1], E.M. Pereira[1], C.A.F. Melo[1], F.A. Coelho[1], A.C.R. Oliveira[1], C. Veloso[1], F.M. Santos[1], N.A.C. Gomes[1], M.S. Monteiro[1], A.P. Poor[2], B.D. Muro[2], R.K.G. Messias[3], and C.A.P. Garbossa*[1], [1]*University of São Paulo, Pirassununga, São Paulo, Brazil*, [2]*Poulpharm, Izegem, West Flanders, Belgium*, [3]*BASF, São Paulo, São Paulo, Brazil*. This study aimed to evaluate the effects of dietary supplementation with a monoglyceride blend (MBLEND; C4:C8:C10) as a replacement for ZnO on growth performance and intestinal health in nursery pigs. A total of 96 piglets (7.08 ± 0.837 kg, 25 days of age) were used in a 42-day trial. Animals were assigned to three treatments (CON: control with no additives, ZnO: zinc oxide, and MBLEND: monoglyceride blend) in a randomized block design. Growth performance (average daily gain, feed intake, and feed conversion ratio) and diarrhea incidence were not affected ($P > 0.05$) by ZnO or MBLEND during all phases. However, MBLEND improved ($P < 0.05$) dry matter, crude protein, and mineral digestibility compared to CON and ZnO. ZnO reduced ($P < 0.05$) IL-1β and tended to reduce ($P < 0.10$) IL-8 cytokine expressions in the jejunal mucosa, whereas MBLEND did not influence these parameters. Villus height, crypt depth, villus:crypt ratio, and intestinal junction gene expression showed no differences ($P > 0.05$) across treatments. ZnO altered beta-diversity ($q < 0.05$), increasing *Tannerellaceae, Bacteroidaceae,* and *Muribaculaceae* families while reducing *Escherichia coli-Shigella* and *Desulfovibrio* genera ($q<0.05$). In conclusion, while ZnO modulated the cecal microbiota and reduced inflammatory cytokines, MBLEND improved nutrient digestibility and reduced harmful bacterial fermentation in the hindgut, presenting positive effects on intestinal health. Both ZnO and MBLEND showed similar results for growth performance and diarrhea incidence. **Key Words:** Gut gene expression, cytokine, microbiome

**58 Regulation of growth and inflammatory responses to lipopolysaccharide challenge in weanling pigs fed dietary nucleotides.** A.S. Lawal*, Y. Fu, M.N. Brackett, O. Adeola, and K.M. Ajuwon, *Purdue University, West Lafayette, IN 47907, USA*. This study was conducted to determine the effects of nucleotide (NL) supplementation in the diets of weanling pigs fed normal crude protein (22% CP, corn-soybean-based) or low crude protein diets (16% CP, corn-soybean-based) on growth performance, nutrient digestibility, and inflammatory responses to lipopolysaccharide (LPS) challenge. Weanling pigs with an initial average body weight (BW) of 7.5 kg were allotted to 4 diets in a 2 × 2 factorial arrangement of CP (normal or low) and NL (not added or added) of 4 pigs/pen and 8 replicates/diet. On d 42, two pigs per pen were intraperitoneally injected with

either saline or LPS (25 μg LPS/kg BW). Rectal temperature (RT) was measured at 0, 2, 3, and 4 h post-challenge, and blood samples were collected at 4 h for analysis. Pigs on normal CP diets had higher ($P < 0.001$) overall ADG, G:F and final BW compared to low CP diets, NL supplementation increased ($P < 0.05$) ADG (d 28–42), final BW, and overall G:F. Supplementation with NL increased ($P < 0.01$) apparent total tract digestibility (ATTD) of dry matter (DM), but low CP diets tended to increase ($P = 0.07$) ATTD of CP. Lipopolysaccharide challenge increased ($P < 0. 001$) RT at 2, 3 and 4 h. Low CP diets decreased ($P < 0.05$) RT at 0 and 4 h while NL supplementation decreased ($P < 0.05$) RT at 4 h post-challenge. Serum glucose and insulin concentrations were decreased ($P < 0.01$ and $P < 0.05$, respectively) by LPS challenge, whereas tumor necrosis factor α (TNF-α) and interleukin-8 (IL-8) concentrations were increased ($P < 0. 001$). Nucleotide supplementation decreased ($P < 0.05$) serum TNF-α concentration but had no effect on IL-8. In conclusion, dietary NL supplementation increased growth performance and ATTD of DM irrespective of CP levels and reduced inflammatory responses in LPS-challenged weanling pigs. **Key Words:** growth performance, lipopolysaccharide, nucleotide

**59 Impact of Dietary Calcium and Microbial Phytase On the Fate of Phosphorus and Calcium in the Gastrointestinal Tract of Growing Pigs.** J. Labarre*[1,2], A. Narcy[3], M. Jlali[4], D.B. Dalot[5], T.D. Crenshaw[6], P. Schlegel[7], and M.P. Létourneau-Montminy[1], [1]*Laval University, Department of Animal Science, Quebec G1V 4G2, Canada*, [2]*Université Paris-Saclay, INRAE, AgroParisTech, UMR MoSAR, 91120, Palaiseau, France*, [3]*INRAE, Université de Tours, BOA, Nouzilly, 37380, France*, [4]*Adisseo France S.A.S, Department of R&I in Monogastric Animal Nutrition, European Laboratory of Innovation, Science and Expertise, 69190 Saint-Fons, France*, [5]*Agriculture and Agri-Food Canada, Sherbrooke R&D Centre, Quebec J1M 0C8, Canada*, [6]*University of Wisconsin, Department of Animal and Dairy Sciences, Madison 53706, USA*, [7]*Agroscope, Swine Research Unit, Posieux, 1725, Switzerland*. Mathematical models of the fate of dietary Ca and P in the gastrointestinal tract (GIT) of growing pigs are in development. However, data are needed to better simulate the effects of Ca and new-generation microbial phytases (PhytM) and their interactions. Thirty-two pigs of 40 kg were used to test the hypothesis that increased dietary Ca will reduce P digestibility by decreasing phytic phosphorus (PP) degradation and P solubility, with a greater impact when PhytM was added. Pigs were allocated to two levels of Ca (0.78 and 0.45%) with or without PhytM (0 vs 500 FTU/kg) while digestible P was set at 0.34% assuming a digestible P and total Ca PhytM equivalency of 0.12%. After 21 days, pigs were euthanized 2.5h after their last meal and digesta samples were collected from various GIT segments. The total P, Ca, PP, and titanium concentrations were analyzed in total and soluble fractions and PP degradation and Ca and P digestibility (disappearance) were calculated. The PhytM increased PP degradation in the stomach ($P < 0.001$) and in the jejunum ($P < 0.001$) and a high Ca reduced PP degradation in the jejunum by 23% ($P = 0.02$), but there was no interaction. The concentration of P was decreased by PhytM in all the GIT segments ($P < 0.001$) and high Ca increased the concentration of P in the jejunum ($P = 0.002$) and the colon ($P = 0.06$). The P concentration in the soluble digesta fraction did not differ among treatments. The lower Ca and PhytM both decreased Ca concentration in all the GIT segments ($P < 0.05$). The high Ca increased the Ca concentration in the soluble digesta fraction in the jejunum ($P = 0.01$) and in the colon ($P = 0.09$). PhytM, but not Ca, increased P digestibility in the jejunum (37 vs 54 %; $P = 0.02$) and colon (49 vs 59%; $P < 0.001$) and Ca digestibility in the colon (38 vs 47 %; $P = 0.05$). These results show that Ca decreases PP degradation without altering the efficiency of PhytM and will be useful for the development of robust P feeding system. **Key Words:** phytase, calcium, phosphorus

**60 Impact of a free organic acid blend on growth performance and mortality of nursery pigs under commercial conditions when combined with pharmacological zinc oxide and free benzoic acid.** A. Hintz*[1], R. Edler[2], E. Little[2], J. A. Acosta[1], B. Lawrence[1], M. Castillo[1], and D. Hancock[1], [1]*NOVUS International, Chesterfield. MO, USA*, [2]*Pipestone Research, Pipestone, MN, USA*. This experiment evaluated the addition of an organic acid blend first 22d (5kg/Ton, ACTIVATE DA Feed Solution, NOVUS International Inc, Chesterfield, MO; OAB) to weaned pigs raised under commercial conditions when used in combination with pharmacological zinc oxide and free benzoic acid. A total of 546 weaned pigs, approximately 21d of age (BW=7.23 kg), were housed in 42 pens and randomly assigned to 1 of 2 treatments (n=21): control (CON), CON+0.5% OAB. The study lasted 49d. In both treatments, a free benzoic acid was included in the diet (3.50, 2.28, and 0.80 kg/Ton for 0-7, 8-14, and 15-22 d, respectively) as was pharmacological zinc oxide (2,500 and 2,000 mg/kg of zinc for d 0-14 and 15-22, respectively) Data were analyzed in SAS V9.4 evaluating growth performance, feed efficiency, mortality, and pig quality. Sub-optimal pigs were visually identified in each pen at d 49 as noticeable small, poor-body condition pigs. The percentage of high-quality pigs were calculated as hundred percent of pigs minus the percentage of sub-optimal pigs at d 49. ADG from 0-49 d was 0.567 kg and 0.559 kg in pigs fed OAB and CON, respectively ($P$ = 0.578). ADFI was 0.859kg and 0.854 kg for pigs fed OAB and those fed CON ($P$ = 0.827). Gain to feed was 0.655 and 0.647 for OAB and CON pigs, respectively ($P$ = 0.391). The final BW was 35.49 kg for OAB and 35.19 kg for CON ($P$ = 0.747). High-quality pigs were 97.80% for pigs fed OAB compared with 95.60% for those in the CON ($P$ = 0.039). Sub-optimal animals were 0.54% for pigs fed OAB and 1.31% for those in the CON ($P$ = 0.285). Mortality for OAB-fed pigs was 1.63%, which tended to be lower than CON 3.11% ($P$ = 0.092). In conclusion, adding OAB on top of a free benzoic and pharmacological zinc oxide improved pig quality at nursery exit and pig survival through the 49-d post-weaning period. **Key Words:** Nursery pigs, intestinal modifiers

**61 Life cycle assessment of low dietary crude protein strategies to improve pig nitrogen efficiency in different geographical contexts.** L Cappelaere*[1], M-P Létourneau-Montminy[1], and F Garcia-Launay[2], [1]*Département des sciences animales, Université Laval, Quebec, Quebec, Canada*, [2]*PEGASE, INRAE, Institut Agro, Saint-Gilles, France*. Dietary crude protein (CP) reduction is a strategy mostly implemented in Europe to improve nitrogen (N) efficiency, protein autonomy and environmental performance. This study aimed to explore the potential of CP reduction to reduce the environmental impacts of pig production in three contrasted contexts (Brazil, Canada, and France), characterized by raw materials used and pig farming practices. For each country, control treatments were formulated in 6 economic contexts to represent current practices and minimum CP reachable was determined. Least cost formulation was used in the six economic contexts to formulate a very low CP diet with a CP level 5 g/kg above minimum CP and an intermediate CP level. Pig performance was simulated with an individual-based model of the pig-fattening unit including InraPorc, and farm gate LCA was performed for all diets. Control CP levels were higher in Canada and Brazil than in France and CP reduction was greater ($P$ < 0.001). Dietary CP reduction increased cereal and free amino acid inclusion in all contexts ($P$ < 0.001), soybean meal inclusion decreased more with CP reduction in the French context ($P$ < 0.001), co-products inclusion increased in Brazilian contexts but decreased in French and Canadian ones ($P$ < 0.001). Growth performance was not affected by CP reduction ($P$ < 0.001). N efficiency was improved in all contexts ($P$ < 0.001), but to a lower extent for the French context, due to higher N efficiency in the control ($P$ < 0.001). Acidification was reduced with CP in all contexts, with larger reductions in Canada and Brazil ($P$ < 0.001). Climate change was only reduced in the French context and increased in other contexts ($P$ < 0.001). Other evaluated impacts were stable or increased ($P$ < 0.001). While acidification is reduced by CP reduction regardless of context, mitigation of other impacts depends on feedstuffs used and especially impact of corn compared to soybean meal in the production context. **Key Words:** low crude protein nitrogen efficiency environmental impacts

**62 Farming conditions and dietary interventions can affect the health and performance of pigs from weaning to growing via modulation of the microbial profile and its metabolism.** D. Luise*[1], G. Palladino[2], F. Correa[1], F. Palumbo[1], M. V. Graziosi[1], E. Perez Calvo[3], G. Litta[3], D. Scicchitano[2], G. Babbi[2], A. Castagnetti[4], S. Rampelli[2], M. Candela[2], P. L. Martelli[2], and P. Trevisi[1], [1]*Department of Agro-Food Sciences and Technologies, University of Bologna, 40127 Bologna, Italy*, [2]*Department of Pharmacy and Biotechnology, University of Bologna, 40126 Bologna, Italy*, [3]*dsm-firmenich, Animal Nutrition and Health, 4303 Kaiseraugst, Switzerland*, [4]*Wellmicro, 40138 Bologna, Italy*. This study aims to evaluate the effect of mix of feed additives on health, performance, fecal microbiome and metabolome of pigs reared in 2 farming conditions. At weaning (T1; 27±2 days of age), 150 pigs (7934 ± 964 g) were divided in 2 farms with an historically high (H) or low (L) performance and antimicrobial misuse and fed either 1) control diet (CO); 2) CO + 5000 mg/kg benzoic acid (BA), 35 mg/kg *E. faecium* and 500 mg/kg muramidase until day 89 (TRT-CO) or 3) as group 2 + BA till d98 (TRT-TRT). Feces and blood were collected on d42 (T2), d80 (T3), and d98 (T4). Feces were analyzed for microbiome (Shotgun) and metabolomic; hemochrome and oxidative stress markers were examined in blood. The H_TRT-TRT had a better ADG T3-T4 ($P < 0.01$) than the H_CO. ADG T1-T4 was higher in the H farm than in the L farm. At T2 and T3, the diet and diet x farm interaction influenced Shannon index ($P < 0.05$); the H_TRT had lowest Shannon index at T2 and highest at T3. At T2 and T3, farm, diet and diet x farm influenced beta diversity ($P = 0.001$), and farm and diet influenced microbial metabolism (KEGG pathways) ($P < 0.05$). At T2, the H farm had an alteration in bile acid and sulfur amino acid metabolism; the L_TRT group had a higher activation of energy metabolism, gut microbiota activity and stress response pathways. At T2, the L group had higher leukocytes, neutrophils, lymphocytes, and monocytes ($P < 0.05$). At T2, L_TRT had a higher biological antioxidant potential (BAP) than H_TRT ($P = 0.05$). At T4, beta diversity was affected by diet, farm and diet x farm ($P < 0.05$); microbial metabolism was affected by diet and diet x farm ($P < 0.05$), but metabolomic did not reveal any difference. L farm had higher neutrophils and lower lymphocytes ($P < 0.01$), and L_CO had higher levels of BAP than all groups in H farm ($P < 0.05$). Overall, this study confirms that both farming conditions and diet can influence pig health and performance, highlighting the robust diet-environment-microbiota-host health interaction. **Key Words:** benzoic acid, leukocytes, muramidase

**63 A water-based organic acid blend improves growth performance, pig quality at nursery exit, and mortality under commercial conditions.** A Hintz*[1], R Edler[2], E Little[2], J Acosta[1], B Lawrence[1], M Castillo[1], and D Hancock[1], [1]*NOVUS International, Chesterfield. MO, USA*, [2]*Pipestone Research, Pipestone, MN, USA*. This experiment evaluated the addition of a water-added organic acid blend the first 14d post-weaning (added to acidify drinking water at 0.12% to a final pH = 4.0, ACTIVATE WD Max Nutritional Feed Acid, NOVUS International Inc, Chesterfield, MO; WAB) under commercial conditions. A total of 546 weaned pigs, approximately 21d of age (BW = 7.23 kg), were housed in 42 pens and randomly assigned to 1 of 2 treatments (n = 21): control (CON) and CON+WAB. This study lasted 49d. A free benzoic acid program (3.50, 2.28, and 0.80kg/Ton for 0-7, 8-14, and 15-22d, respectively) and pharmacological zinc oxide (2,500 and 2,000mg/kg of zinc oxide for d 0-14 and 15-22, respectively) were part of the nursery feeding program. Data were analyzed in SAS V9.4, evaluating growth performance, feed efficiency, mortality, and pig quality. Sub-optimal pigs were visually identified in each pen at d 49 as noticeable small, poor-body condition pigs. The percentage of high-quality pigs were calculated as hundred percent of pigs at d 49 minus the percentage of sub-optimal pigs. ADG from 0-49 d was 0.575 kg and 0.559 kg in pigs fed WAB and CON, respectively ($P = 0.045$). ADFI was 0.883 kg for pigs fed WAB and 0.854kg for those fed CON ($P = 0.215$). Gain to feed was 0.651 and 0.647 for WAB and CON pigs, respectively ($P = 0.748$). The final BW was 35.93kg for WAB and 35.19kg for CON ($P = 0.318$). High-quality pigs were 98.88% for pigs fed WAB compared with 95.60% for those fed CON ($P = 0.005$). Sub-optimal pigs were 0.38% for those fed WAB, which was lower than the 1.31% observed in the CON ($P = 0.221$) group. Mortality for WAB-fed pigs was 0.74%, which tended to be lower than the 3.11% observed in CON pigs ($P = 0.008$). In conclusion, adding WAB on top of free benzoic acid and pharmacological zinc oxide resulted in greater weight

gain, better pig quality at the end of the nursery, and decreased mortality. **Key Words:** Nursery pigs, intestinal modifiers

**64 Effects of supplemented guanidinoacetic acid on animal-related indicators of sows and their offspring.** S. J. Esfarjani N.*[1,2], P. Loibl[3], J. Steinhoff-Wagner[1], and G. Dusel[2], [1]*Technical University of Munich, Freising-Weihenstephan, Germany*, [2]*University of Applied Sciences Bingen, Bingen am Rhein, Germany*, [3]*Alzchem Trostberg GmbH, Trostberg, Germany*. Guanidinoacetic acid (GAA) is the direct precursor of creatine and is often used as a creatine source in feed. Due to the limitation of creatine synthesis in the body, supplementation of GAA to the diet may lead to higher creatine concentrations in sows' milk which potentially enhance piglet growth and reduce piglet mortality. In addition, energy efficiency for productivity of sows with high energy requirements might be improved. The aim of this study was to evaluate if GAA supplementation will improve the reproductive performance of sows, the piglet birth weight and weight gain after birth. A total of 93 sows (Topigs TN70) were allocated to two feeding groups (GAA, n = 44 and control, n = 49) based on three parity classes (I: 0, n = 18; II: 1-4, n = 45; III: 5-13, n=30). GAA was supplemented at 0.10 % to the feeds of the trial group. Sows were observed from gestation (d 0-115) and lactation phase (d 115-143). Body weight (BW), backfat thickness (BFT), body condition score (BCS), farrowing and litter performance as well as birth and weaning weight of the piglets were recorded. In the ANOVA (SAS 9.4), feeding group and parity were considered as fixed effects and individual sow as random effect. Sows' performance parameters, feed intake, milk yield, total number of piglets born and born alive, litter weight at birth and weaning, and the number of weaned piglets were comparable between the feeding groups. Piglet birth weight in parity class II was higher in the GAA group compared to the control group (LSM ± SE: 1.36 ± 0.07 kg vs. 1.27 ± 0.07 kg; $P < 0.02$). Following up, the weaned piglets in parity class II were heavier in the GAA group (7.18 ± 0.27 kg vs. 6.76 ± 0.28 kg; $P < 0.05$). There was an interaction between the treatment and parity in the birth weight ($P < 0.001$) and weaned weight of the piglets (P = 0.02). In conclusion, it seemed that the birth weight of piglets increased with the addition of GAA to sows' diets which likely resulted in increased weaning weight. **Key Words:** Sow, GAA (guanidinoacetic acid), reproductive performance

**65 Non-nutritive sweeteners induce unique metabolomic changes in weaned pigs compared to antibiotic supplementation.** Kwangwook Kim*, *Michigan State University, East Lansing, Michigan, USA*. Our previous results demonstrated that supplementation with non-nutritive sweeteners (NNS) improved growth performance and reduced diarrhea in weaned pigs. This study aimed to further investigate the effects of NNS on the colon digesta metabolomic profiles of weaned pigs, compared to in-feed antibiotics. A total of 288 pigs (6.21 ± 0.45 kg, 21 ± 1 day old) were randomly assigned to one of four dietary treatments (12 pigs/treatments): negative control (NC; basal nursery diet), positive control (PC; NC + 50 mg/kg antibiotic), sucralose (SCL; NC + 150 mg/kg sucralose), and neotame (NEO; NC + 30 mg/kg neotame). Pigs were fed the diets for 28 days post-weaning, split into Phase 1 (days 0 to 14) and Phase 2 (days 15 to 28). On days 14 and 28, 6 pigs/group were euthanized to collect colon digesta for metabolomic analysis using gas chromatography time-of-flight mass spectrometry. Data were statistically analyzed (fold change > 2.0; $P < 0.05$) and evaluated using online MetaboAnalyst tool. On day 14, NNS reduced 16 metabolites and increased levels of 1,5-anhydroglucitol and cadaverine compared to NC, primarily affecting amino acid metabolism. By day 28, SCL and NEO supplementation resulted in reductions in 6 metabolites, including 1-monoolein, inositol-4-monophosphate, inosine, oleic acid, serotonin, and uridine, relative to NC, indicating effects on energy and lipid metabolism. When compared to PC, the NNS groups showed increased levels of 13 metabolites and reduced levels of linoleic acid, oleic acid, 1-monoolein, and linolenic acid, with these changes linked to carbohydrate metabolism, fatty acid biosynthesis, and DNA and RNA synthesis pathways. In conclusion, supplementation with SCL and NEO distinctly altered colon digesta metabolite levels and pathways associated with nutrient metabolism, demonstrating mechanisms that differ from those of in-feed antibiotics. These findings offer a more comprehensive understanding of the underlying mechanisms of NNS. **Key Words:** non-nutritive sweeteners, colon digesta metabolomics, weaned pig

**66 Monosaccharides degradation and in vitro fermentation dynamics of total dietary fiber from cereal-based feed ingredients for growing pigs.** Y. W. Xu*, M. Y. Huang, Y. Cao, J. B. Zhao, D. D. Han, and J. J. Wang, *College of Animal Science and Technology, China Agricultural University, Beijing, China*. To support the development and effectiveness of new pig feed resources, this study investigated the monosaccharide composition of total dietary fiber (TDF) from rice bran meal, wheat bran, corn bran, corn DDGS, oat bran, and barley bran, and evaluated their bioavailability using a pig-derived microbiome. *In vitro* fecal microbial fermentation (n=6) was conducted to assess pH changes, gas and short chain fatty acids (SCFAs) production. The monosaccharide composition of TDFs was detected using ion chromatography and the data were analyzed via one-way ANOVA with SPSS 20.0. Duncan's multiple comparison test was applied when significant difference ($P < 0.05$) was observed. Compared to other TDFs, oat bran exhibited the greatest degradability of total monosaccharides, xylose, and glucose; while rice bran meal showed the lowest degradability of total monosaccharides and glucose ($P < 0.05$). Fermentation dynamics showed that oat bran had the highest fermentability (producing the most SCFAs and gas), but the slowest fermentation speed. In contrast, rice bran meal displayed lower fermentability, but a faster fermentation speed ($P < 0.05$). Further analysis revealed that acetate was the main product, with elevated levels of propionate and lactate observed in the early fermentation (8 h), followed by a rise in acetate in the plateau phase (40 h). Oat bran produced more butyrate than rice bran meal, whereas rice bran meal produced the least acetate among the TDFs ($P < 0.05$). Spearman rank correlation showed significant positive correlations between monosaccharides (glucose and xylose) degradability and SCFAs production (acetate and propionate). In short, oat bran shows strong potential for feed development due to its high fermentability and beneficial metabolite, whereas rice bran meal should be used with caution due to its low fermentability. Future studies should focus on the impact of specific polysaccharides, particularly glucan and xylan, on the fermentation dynamics of cereal fibers. **Key Words:** cereal fibers, fermentation dynamics, monosaccharides degradability

**67 Efficacy of combined feed additive on phosphorus digestibility and retention in rapeseed cake and rapeseed meal when fed to growing pigs.** I. K. Matthiesen*[1,2], J. V. Nørgaard[2], L. H. B. Hansen[3], T. Hinrichsen[4], B. Fisker[4], and M. E. van der Heide[2], [1]*Danish Agro, Karise, Denmark*, [2]*Aarhus University, Tjele, Denmark*, [3]*Novonesis, Kongens Lyngby, Denmark*, [4]*DSM-Firmenich, Broendby, Denmark*. This study evaluated apparent total tract digestibility (ATTD) of dry matter (DM), organic matter (OM) and phosphorous (P), and retention of P in G/F pigs fed rapeseed cake (RC) and rapeseed meal (RM) supplemented with a combined feed additive. Four dietary treatments with rapeseed as sole protein source were formulated to target 15.2% crude protein: RC, RC with additive (RC+), RM, RM with additive (RM+). The additive contained $1.50x10^9$ CFU *Bacillus amyloliquefaciens*, 2000 FTU phytase and 270 IU xylanase/kg feed. Monocalcium phosphate was not used in the feed to only test P digestibility of rapeseed and the effect of additive. The RC and RM diets contained 5.64 g and 5.09 g P/kg DM, respectively. Treatments were tested during two balanced periods of 21 days with 8 pigs/diet (n = 32). Pigs with average BW of 42 kg were placed individually in single pens with ad libitum access to feed and water. At day 10, pigs were placed into balance cages and fed 800 g feed twice/day. After 5 days, they were fitted with catheters. Feces, urine and feed residues were collected and weighed daily for 7 days and pooled for each pig. Data were analysed by ANOVA linear model procedure with additive and rapeseed type as fixed effects and period as random effect. Post-hoc analysis was made with emmeans (R v4.4.1). The additive improved ATTD of P ($P < 0.01$; RC: 28.7% vs RC+: 53.3%; RM: 27.5% vs RM+: 52.2%) but did not affect ATTD of DM ($P = 0.30$; RC: 83.2% vs RC+: 83.5%; RM: 83.9% vs RM+: 84.3%) or OM ($P = 0.93$; RC: 85.4% vs RC+: 85.4%; RM: 86.5% vs RM+: 86.5%). The retention of P increased ($P < 0.01$; RC: 2.11 vs RC+: 3.06; RM: 2.00 vs RM+: 2.95 g/day) also relative to P intake ($P < 0.01$; RC: 0.27 vs RC+: 0.40; RM: 0.29.00 vs RM+: 0.42). In conclusion, the additive significantly increased ATTD of P with 85.7% for RC and 89.8% for RM and increased total retention of P with 45.0% and 47.5%, respectively. This indicates that the phytase in the additive increased the digestibility of P in both rapeseed sources. **Key Words:** Rapeseed, Apparent total tract digestibility, Phosphorous retention

**68 Effects of Three Probiotic Preparations on Growth Performance, Oxidative Stress, and Gut Microbiota of Duroc-Landrace-Yorkshire Ternary Hybrid Growing Pigs.** T Kiros*[1], H Zhang[2], S XU[2], X Shen[3], and Z YU[3], [1]*Phileo by Lesaffre-North America, Milwaukee, WI, USA*, [2]*Phileo by Lesaffre- China, Shanghai, China*, [3]*Nanjing Agricultural University, Nanjing, China*. Probiotics has been shown to improve gut health, nutrient digestibility, and growth performance in pigs. This study investigated the effects of Yeast and Bacillus-based probiotics on the health and performance of growing pigs. A total of 240 weaned piglets (10.0±1 kg) were randomly assigned to one of four treatment groups (12 pens/treatment, 5 pigs/pen): 1) Basal diet (Corn-soybean meal based diet, control), 2) basal diet + *Saccharomyces cerevisiae*, Sc 47 yeast (1 kg/MT, 0-30 days, and 0.5 kg/MT, 30-90 days post weaning; Yeast), 3) basal diet + Bacillus mix 1 (0.5 kg/MT; BacM1), or 4) basal diet + Bacillus mix 2 (0.5 kg/MT; BacM2). BacM1 contained three strains of *B. velezensis* and one strain *B. subtilis*, while BacM2 contained *B. licheniformis, B. pumilus, B. amyloliquefaciens* mixed in equal proportion to make 2x10^9 cfu/g. Performance was measured on days 0, 30, 60, and 90. Fecal and blood samples were collected on day 90 for microbiota and oxidative stress analyses. Pigs fed BacM1 showed a trend ($P = 0.06$) for higher average daily gain, resulting in a significantly lower ($P < 0.05$) FCR compared to control from day 30 onward. Total antioxidant capacity was higher ($P < 0.01$) in the Yeast and BacM1 groups compared to the other two groups. Superoxide dismutase was higher, and malondialdehyde lower in all probiotic groups versus control ($P < 0.01$). Protein digestibility increased for Yeast and BacM1 groups ($P < 0.01$), while it showed a trend for BacM2. Total short-chain fatty acids were higher in BacM2 ($P < 0.01$) and BacM1 ($P < 0.05$), with propionic acid also higher in these two groups ($P < 0.05$) compared to the other two groups, while acetic acid was significantly higher ($P < 0.01$) in all the supplemented groups. In conclusion, supplementation of pig diets with Yeast and Bacillus probiotics decreased oxidative stress, increased SCFA, mainly acetic acid, and improved protein digestibility. The BacM1 showed a trend for improved weight gain and significant increase in feed efficiency. **Key Words:** Gut health, Pigs, Probiotics

**69 Feeding live black soldier fly larvae increases the voluntary feed intake of suckling piglets.** N.S. Stöhr*[1,2], L Schneider[1], J Stracke[2], R Jha[3], and G Dusel[1], [1]*University of Applied Sciences Bingen, Bingen am Rhein, RLP, Germany*, [2]*University of Bonn, Bonn, NRW, Germany*, [3]*University of Hawaii, Manoa, HI, USA*. Piglets naturally adapt to solid feedstuff (plants, nuts, insects) over several months, whereas commercial piglets are weaned abruptly at 3-4 weeks of age. Early solid feed exposure is essential to prepare their digestion (enzyme training) for the post-weaning period. Live black soldier fly larvae (LBSFL) might be a potential strategy for piglets used to liquid milk, as they are low in dry matter (DM) (< 30%) and high in protein (~50% in DM) and fat (~25% in DM) content. In this study, a total of 599 piglets (average initial body weight of 1.58kg and age of 1.5 days) from 43 sows (average of 14 piglets/litter) across two experimental runs were equally allocated into two treatments: CON = creep feed (CRF), LAR = CFR + LBSFL. Piglets were weighed at trial start and weekly, on days 7, 14, 21, and 28, to evaluate growth performance. LBSFL and CRF were offered in separate troughs to record feed intake. Each litter had access to two troughs (CON: 2× CRF, LAR: 1× CRF + 1× LBSFL) to ensure equal feeding space availability. CRF and LBSFL were weighed daily and offered fresh each day. If a feed trough was empty the next day, the amount of CRF or LBSFL was increased to simulate *ad libitum* feeding. Data were analyzed using one-way-ANOVA (SPSS, IBM). LBSFL feeding did not affect the weaning weight of the piglets at day 28 ($P > 0.05$). Piglets fed LBSFL tended to exhibit a 32% higher average daily CRF intake (ADFI) compared to CON during the whole suckling period ($P = 0.057$), while CRF consumption was 42% higher in the last week before weaning ($P = 0.022$). In addition, larvae feeding resulted in a 77% increase in total DM intake (CRF incl. LBSFL on DM basis) during the whole suckling period ($P < 0.001$). This led to an increase in overall nutrient intake, especially lysine (+56%, $P < 0.001$) and metabolizable energy (+91%, $P < 0.001$). In conclusion, LBSFL feeding promotes overall voluntary feed intake during the suckling period and enables the uptake of a diverse range of feedstuffs. **Key Words:** Insect, Hermetia illucens, pre-weaning

**70 Impact of a feed additive containing a Bacillus-based probiotic, microbial phytase and carbohydrase on protein and amino acid digestibility in rapeseed cake and rapeseed meal for growing-finishing pigs.** I. K. Matthiesen*[1,2], J. V. Nørgaard[2], T. Hinrichsen[3], B. Fisker[3], and M. E. van der Heide[2], [1]*Danish Agro, Karise, Denmark*, [2]*Aarhus University, Tjele, Denmark*, [3]*DSM-Firmenich, Broendby, Denmark*. This study examined apparent ileal digestibility (AID) of crude protein (CP), dry matter (DM) and total amino acids (TAA), and apparent total tract digestibility (ATTD) of CP and DM in rapeseed cake (RC) and rapeseed meal (RM) when supplemented with a combined feed additive as an alternative to soy in feed for G/F pigs. Four dietary treatments were used in a 2x2 factorial design with rapeseed as sole protein source targeting 15.2% CP using titanium oxide as marker: RC, RC with additive (RC+), RM, RM with additive (RM+). Diets with additive contained 1.50 x $10^9$ CFU *Bacillus amyloliquefaciens*, 2000 FTU phytase and 270 IU xylanase/kg feed. Forty crossbred pigs (32.2±3.11 kg) were randomly allotted to the 4 treatments. Pigs were placed in group pens and offered a standard feed ad libitum the first 7 days. At day 8, the treatments started with semi-restrictive feeding. Pigs were slaughtered after 14 days on treatment and a spot sample of feces and digesta from the last 100 cm of the terminal ileum were collected and freeze dried for nutrient analyses. Data were analysed by ANOVA using a linear model procedure with additive and rapeseed type as fixed effects. Post-hoc analysis was made with the emmeans package (R v4.4.1). Addition of the feed additive decreased AID of CP for both rapeseed cake and meal ($P$ = 0.05; RC: 66.8% vs RC+: 62.0%; RM: 60.5% vs RM+: 55.8%) and tended to decrease AID of TAA ($P$ = 0.09; RC: 69.7% vs RC+: 65.4%; RM: 64.4% vs RM+: 60.2%). No effect was seen in AID of DM ($P$ = 0.52; RC: 95.7% vs RC+: 95.4%; RM: 95.8% vs RM+: 95.6%). The additive improved ATTD of DM ($P$ < 0.01; RC: 92.38% vs RC+: 93.52%; RM: 93.35% vs RM+: 93.50%), but did not affect ATTD of CP ($P$ = 0.47; RC: 73.8% vs RC+: 72.9%; RM: 71.0% vs RM+: 70.2%). In conclusion, AID and ATTD of CP and DM did not show consistent results. **Key Words:** Rapeseed, Apparent ileal digestibility, Apparent total tract digestibility

**71 Microencapsulated organic acids and essential oils enhance sow performance and piglet outcomes in field conditions.** O.O Babatunde*[1], G Tactacan[1], L Lahaye[1], A Seemacharoensri[1], and P Assavacheep[2], [1]*Jefo Nutrition Inc., St-Hyacinthe, QC, Canada*, [2]*Chulalongkorn University, Bangkok, Thailand*. Reducing antibiotic use in sow diets necessitates effective strategies to protect intestinal health during the reproductive cycle, mitigate stress-induced body condition loss, and enhance productive performance. Organic acids and essential oils (OA+EO) possess antimicrobial properties, and when microencapsulated, may improve intestinal health and nutrient utilization. This study aimed at investigating the effects of a microencapsulated blend of OA+EO [P(OA+EO)] on sow performance and piglet outcomes during lactation. A total of 322 farrowing sows (Large white x Landrace; average parity = 3.06 ± 1.95) were randomly assigned to two dietary treatments: a control group receiving a standard lactation diet and a treated group supplemented with P(OA+EO) at 10 g/sow/day on top of feed for 35 days (from 7 days pre-farrowing to weaning at 28 days). Data were analyzed using Student t-test and paired t-test with significance set at $P$ < 0.05. Sows fed P(OA+EO) exhibited significantly reduced ($P$ < 0.05) body condition score loss, backfat loss, and shoulder ulcer incidence compared to the control group. In addition, sows fed P(OA+EO) had higher ($P$ < 0.05) feed intake than sows in the control group. Piglets nursing from P(OA+EO)-fed sows had a 15% reduction ($P$ < 0.05) in diarrhea incidence and a 5% decrease ($P$ < 0.05) in pre-weaning mortality. Additionally, P(OA+EO) supplementation increased ($P$ < 0.05) the number of weaned piglets per litter (11.8 vs 11.3) and improved piglet weaning weight (6.11 vs. 5.90 kg). Average daily litter weight gain improved ($P$ < 0.05) by 9% in the treated group vs. the control group (1.96 vs 1.80 kg/day). In conclusion, the microencapsulated OA+EO blend reduced diarrhea incidence in piglets, farrowing-related body condition loss, and improved sow and piglet performance. These findings highlight the potential of functional feed additives to promote performance, digestive function and nutrient efficiency, supporting the transition away from antibiotic use. **Key Words:** microencapsulated organic acids and essential oils, piglet performance, sow performance

**72 Effects of supplemental phytase on growth performance, blood inositol levels, and immune characteristics of weanling pigs.** A Mallea*[1], SA Lee[2], and H Stein[1,2], [1]*Division of Nutritional Sciences, University of Illinois at Urbana-Champaign, Urbana, IL, USA*, [2]*Department of Animal Sciences, University of Illinois at Urbana-Champaign, Urbana, IL, USA*. The objective was to test the hypothesis that weanling pigs need microbial phytase in diets to be able to maintain pre-weaning levels of plasma inositol throughout the post-weaning phase. The second hypothesis was that phytase improves growth performance and immune characteristics of weanling pigs. A control diet contained Ca and P at the requirements of pigs. Two additional diets contained 1,000 or 2,000 units of phytase (FTU) and 0.08% digestible P and 0.12% total Ca less than the requirements to account for the expected releases of P and Ca by phytase. A total of 150 newly weaned pigs [initial body weight (BW): 5.43 ± 0.43 kg] were randomly allotted to the 3 diets. There were 10 pen replicates per treatment and 5 pigs per pen. Blood sample was collected from 2 pigs per pen on the day before weaning, and on day 14, 28, and 42 post-weaning. Concentrations of tumor necrosis factor-alpha (TNF-α) and inositol were analyzed. The statistical model included diet as fixed variable and replicate as random variable. Overall average daily gain of pigs fed the 2 phytase diets tended to be greater ($P < 0.10$) compared with pigs fed the control diet, but average feed intake was not affected by dietary treatment. Overall gain:feed of pigs fed the 2,000 FTU diet was the greatest ($P < 0.05$), followed by the 1,000 FTU diet ($P < 0.05$) and the control diet ($P < 0.05$). Pigs fed the 2 phytase diets had greater ($P < 0.05$) plasma inositol on day 14 and 28 than pigs fed the control diet. On day 28 pigs fed the 2 phytase diets had reduced ($P < 0.05$) plasma TNF-α compared with pigs fed the control diet. In conclusion, pre-weaning levels of plasma inositol were maintained in pigs fed diets containing phytase during the initial 42 days post-weaning. Supplementation of phytase to diets that contained P and Ca below the requirements for weanling pigs improved growth performance and reduced plasma TNF-α, which indicates that phytase had positive effects on modulating inflammatory response. **Key Words:** inositol, phytase, weanling pigs

**73 Improving creep-feed consumption with an intake promoter enhances performance in post-weaning piglets.** A.J. Galindo[1,3], D. Solà-oriol*[1], F. J. Crespo[2], and M. Paniagua[3], [1]*Animal Nutrition and Welfare Service, Bellaterra, Catalunya, Spain*, [2]*HTBA (HealthTech Bio Actives, S.L.U.), Barcelona, Catalunya, Spain*, [3]*Quimidroga SA, Barcelona, Catalunya, Spain*. Creep feeding is a key management practice for improving post-weaning (PW) performance and health by familiarizing piglets with solid feed and enhancing digestive system maturation. Thus, this trial aimed to evaluate the effects of supplementing an intake promoter containing volatile compounds based on sow's placenta, colostrum and milk with Neohesperidin DC in lactation and pre-starter diets in piglets. A total of 513 mixed sex suckling piglets (7±1.49 days old; 2.21±0.02 kg BW) from 40 litters ([Landrace x Yorkshire] x Pietrain) were assigned to one of two treatments: a basal diet (CON) or CON supplemented with the intake promoter (EXP) in the creep-feed and pre-starter concentrate. During lactation (LT: from day 7 to weaning), creep-feed diets included indigo carmine as a feed consumption fecal marker. At weaning (27.6 ±1.53 days old), identification of eater piglets through rectal swab was performed, and 66 piglets per treatment were randomly selected and housed in 12 pens to continue the trial in pre-starter (PS: from weaning to 14 days PW) and starter phase (ST: from PS to 35 days PW). BW and FI were recorded at 7, 28, 42, and 63 days of life and ADG, ADFI and FCR were calculated for each period. Data were analyzed using one-way ANOVA with R, considering treatment as a fixed effect. Eater proportion was greater ($P < 0.001$) in EXP (24,8%) group than in CON (8,3%) during LT. Also, ADFI was higher ($P < 0.05$) for EXP litters (20.0g/day) compared to CON (15.3g/day) in this phase. In PS, FCR was higher ($P < 0.05$) in EXP group. ST ADG diverged ($P < 0.001$) between EXP (318 g/day) and CON (273g/day). At the end of the study, pigs fed EXP treatment achieved higher ($P < 0.05$) PW ADG (274 vs. 255g/day) and final BW (16.12 vs. 15.30kg) demonstrating a carry-over effect of the intake promoter. In conclusion, adding the intake promoter on LT and PS diets enhanced litter LT feed intake, number of creep-feed consumers and growth performance during starter phase. **Key Words:** creep-feed, piglets, weaning

**74 Effects of reducing calcium-phosphorus ratio and adding phytase and 25-Hydroxyvitamin D3 on growing-finishing pigs performance, digestibility and bone characteristics.** F. M. Santos[1], C. C. S. Martins*[2], E. Perez-Clavo[3], M. F. Prata[2], C. P. Lozano[2], and C. A. P. Garbossa[1], [1]*University of São Paulo, Pirassununga, SP, Brazil*, [2]*DSM-Firmenich, São Paulo, SP, Brazil*, [3]*DSM-Firmenich, Kaiseraugst, Switzerland*. Calcium (Ca) and Phosphorus (P) requirement is important for bone mineralization and muscle deposition. Excessive Ca reduces P digestibility, increases P excretion and consequently increases environmental pollution. Phytase and vitamin D are involved on mineral metabolism which could fine-tune mineral utilization allowing Ca reduction without compromising growth performance and bone health. 144 pigs (PIC, 28.26 kg BW) were used to evaluate the effects of reducing Ca:P ratio with or without 25(OH)$D_3$ supplementation, on performance, nutrients digestibility and bone characteristics for 91 days. Pigs were distributed in 36 pens (4pigs/pen) with 3 treatments: T1: Control – 2:1 Ca:P ratio; T2: 1.4:1 Ca:P ratio; T3: T2 + 25(OH)$D_3$ at 50mcg/kg. Four feeding phases (G1, G2, F1 and F2) of corn-soybean meal-based diets with phytase (600 FYT/kg) and variable vitamin D3 levels (G1 and G2 = 1,500; F1 = 1125 and F2 = 750 IU/kg) were offered ad libitum. Body weight (BW), average daily gain (ADG), feed intake (AFI), and feed conversion ratio (FCR) were analyzed. The apparent total tract digestibility (ATTD) of nutrients was measured at the end of G1 phase. At d91, the third metacarpal was collected to analyze the mineral content (BMC) and bone breaking strength (BBS). All data were analyzed using SAS software to compare the means by Tukey test ($P < 0.05$). Results showed that low Ca:P ratio (T2) reduced significantly ADG in all the phases and BMC and BBS ($P < 0.001$) compared to control (T1) at the end of the trial. The inclusion of 25(OH)$D_3$ at low Ca:P ratio (T3) compensated for loss of ADG with similar results as T1 and improved the ATTD of Ca making mineral absorption more efficient. Moreover, while 25(OH)$D_3$ was not able to compensate for reduction of BMC and BBS, the content of Ca and P at the bone tended to be increased ($P < 0.1$). In conclusion, 25(OH)$D_3$ supplementation in reduced Ca:P ratio diets mitigated the negative effects on growth performance by improving mineral absorption. **Key Words:** vitamin D, mineral nutrition, 25(OH)D3

**75 Different weaning ages and Zinc sources influence gut health in nursery pigs.** P. Trevisi*[1], D. Luise[1], C. Negrini[1], F. Correa[1], M. Mazzoni[2], A. Romeo[3], Y. J. Manaig[3], and A. Monteiro[3], [1]*Department of Agricultural and Food Sciences, 40127 Bologna, Italy*, [2]*Department of Veterinary Medicine, University of Bologna, 40064 Ozzano dell'Emilia, Italy*, [3]*Animine, 74960 Annecy, France*. This trial aimed to assess gut health and microbial composition in piglets of different weaning ages. At weaning, 96 pigs were arranged in a 2x3 factorial design that included two weaning ages: Early (E, 21 days) and Normal (N, 26 days), and ~ 120 mg/kg of Zn from 3 sources: Zn sulphate (CO), Zn Glycinate (Gly) and potentiated Zinc Oxide (HiZox®, Hi). Piglets were divided into 8 replicates (2 piglets/replicate) balanced by litter of origin and body weight. Piglets were weighted at weaning (d0) and weekly until d21, with slaughter points at d8 (one piglet/replicate) and d21 (remaining piglets), where jejunum content and tissue were collected for pH measurement, morphology, microbiota, and lactic acid concentration. Data were analysed using a linear mixed model and ANOVA, with diet, weaning age, and their interaction as fixed factors, and litter of origin as random factor. Results indicated a trend for diet influencing the jejunum pH at d8 ($P = 0.08$), where the Hi piglets showed lower pH. The EHi group had lower jejunum pH at d8 compared to EGly ($P = 0.04$). In the same timepoint, a tendency for interaction between weaning age and diet was observed for lactate concentration ($P = 0.07$), with EHi showed a tendency to increase lactate compared to EGly ($P = 0.05$). Villus height and villus height-to-crypt depth ratio (VH:CD) tended to be influenced by the weaning age at d8, resulting higher values in the N group ($P < 0.10$). At d21, crypt depth was lower in EHi than ECO ($P = 0.02$) and VH:CD was higher in EHi compared to ECO ($P = 0.04$). Regarding microbiota, the EHi group showed a higher abundance of HT002, belonging to the *Lactobacillaceae* family ($P = 0.04$) at d8. At d21, EHi piglets were characterized by a higher abundance of ASV 43 associated to the genus *Weissella* ($P = 0.046$). In conclusion, the use of potentiated ZnO may contribute to reduce the detrimental effects of weaning on gut homeostasis. **Key Words:** Micronutrients, Minerals, Gut microbiota

**76 Impact of dietary tributyrin in late gestation and lactation diets on litter performance and immune status of prolific sows.** John K. Htoo*[1] and Sarah Asmussen[2], [1]*Evonik Operations GmbH, Hanau-Wolfgang, Germany*, [2]*Livestock feed tests Denmark ApS, Bylderup Bov, Denmark*. Research showed that butyrate supplementation during gestation and lactation can increase litter weight gain. A study was conducted to evaluate the effects of supplementation with a tributyrin (TB) in sow diets during the last 21 days of gestation throughout lactation on immune status and performance of sows and suckling piglets. Ninety sows (Topigs x DanAvl Duroc; 45 sows (parity 2 to 7) per treatment) were allotted to 2 treatments: 1) a lactation diet based on wheat, barley, soybean meal in pellet (0.90% SID Lys, 2,311 kcal/kg NE; Control) and 2) a tributyrin was supplemented on-top to the basal diet (1 kg/ton). Ten sows were selected per treatment to collect colostrum on day 1 of lactation and blood samples (on day 3 lactation) to analyze serum concentrations of tumor necrosis factor alpha (TNF-α) and superoxide dismutase (SOD). Sow body condition scores, litter size and piglet body weight (BW), and pre-weaned mortality were recorded. On day 1 of lactation, the litter size was equalized at 15 piglets. Data were analyzed by ANOVA using Proc GLM of SAS. The model included parity as covariate. Tributyrin supplementation tended to increase ($P$ = 0.08) the overall sow feed intake and reduced ($P$ = 0.005) backfat depth loss during lactation, however, sow BW and body condition scores, the number born alive, weaned pigs per sow and piglet mortality were not affected. Supplementation with tributyrin tended to increase litter weight gain compared with Control (88.4 vs. 83.7 kg; $P$ = 0.08), however, it did not affect serum concentrations of TNF-α (200 vs. 264 pg/mL; $P$ = 0.331) and serum SOD (8.4 vs. 10.3 U/ml; $P$ = 0.449) and Brix value as indicative of IgG content in milk (23.3 vs. 21.7%; $P$ = 0.485) compared with the control. Overall, supplementation with a tributyrin in sow diet increases litter weight gain at weaning and reduces backfat depth loss which indicates that tributyrin may increase body energy reserves for sows. **Key Words:** piglets, sows, tributyrin

**77 Effect of encapsulated amino acids on the growth and faecal metagenome of nursery pigs.** F. Correa*[1], D. Luise[1], F. Palumbo[1], A. Simongiovanni[2], S. Lecuelle[2], A. Castagnetti[3], and P. Trevisi[1], [1]*Department of Agricultural and Food Sciences, University of Bologna, 40127 Bologna,Italy*, [2]*EUROLYSINE, 80080 Amiens, France*, [3]*Wellmicro srl, 40128 Bologna, Italy*. Nursery pigs encounter various challenges that can significantly affect their growth and gut health. Amino acids (AAs), serving as precursors for energy and functional molecules as well as gut microbiota modulators, can support and restore gut health. However, for free AAs to effectively reach the hind-gut microbiota, they must be protected against degradation in the stomach environment. This study examined the effects of encapsulated AAs on growth, gut health, and microbiome in piglets. A total of 168 weaned pig (28 days), were assigned to seven dietary groups: a Control group (standard nursery diet), and six groups receiving either lysine (Lys, 2 kg/t), tryptophan (Trp, 2 kg/t), branched-chain AAs (BCAA, 2 kg/t), or combinations of Lys (1 kg/t) + Trp (1 kg/t), Lys (1 kg/t) + BCAA (1 kg/t), or Trp (1 kg/t) + BCAA (1 kg/t) on top of the basal diet. Body weight (BW), average daily gain (ADG), feed intake (FI), and feed-to-gain ratio (F:G) were recorded weekly. At day 14 and 28, free AAs levels in ileum content and faeces were quantified (HPLP analysis), while caecum content underwent metagenomic analysis. Data were analyzed using a linear mixed model with the experimental group as a fixed effect and pen as a random effect. Although BW and FI showed no significant differences, ADG and F:G improved between days 21–28 in piglets receiving encapsulated BCAA or Trp+BCAA vs. Control ($P$ < 0.05). Free Trp was higher in the ileum of encapsulated Trp group compared to the Control ($P$ < 0.05), but Trp disappearance (Trp ileum – Trp faeces) did not differ between groups, confirming that AAs were targeted released and utilized in the colon. Metagenomic analysis showed higher caecal Kahnovirus with Trp, Flintibacter sp. KGMB00164 with BCAA, and Roseburia hominis with Trp+BCAA (LDA>3, $P$ < 0.05). These results highlight the efficacy of encapsulated AAs, particularly BCAA and Trp combinations, in improving growth performance, nutrient utilization and modulated the gut microbial ecosystem. **Key Words:** functional amino acids, microbial functions

**78 Impact of benzoic acid source either free or using fat matrix protection on weaned pig response to diarrhea under an *E. coli* F18 challenge, and on growth performance and feed efficiency in commercial conditions.** J. A. Acosta*, B. Lawrence, A. Hintz, M. Castillo, and D. Hancock, *NOVUS International, Chesterfield. MO, USA*. Two experiments were conducted to evaluate the addition of free benzoic acid (5.0kg/Ton, Vevovitall, DSM Nutritional Products, Parsippany, NJ; FBA) or a fat matrix-protected benzoic acid (2.5kg/Ton, NOVUS PB Feed Solution, Novus International Inc, Chesterfield, MO; PBA) to pigs challenged with F18 *E. coli* or raised under commercial conditions. In Exp 1, 72 weaned barrows (BW=5.57kg) were housed in 24 pens and randomly assigned to 1 of 4 treatments (n=6): non-challenged control (NCC), challenged control (CC), CC+FBA, and CC+PBA. After 10-d of acclimation, challenged pigs were infected with F18 E. coli and allowed to recover for 11-d post-infection (dpi). In Exp 2, 1,134 weaned pigs (BW=5.60kg) were housed in 42 pens and randomly assigned to 1 of 3 treatments (n=14): control (CON), CON+FBA, and CON+PBA. This study lasted 21d. In both experiments, the diets had 1,500mg/kg of zinc as zinc oxide. Data were analyzed in SAS V9.4 using pre-planned contrasts. In Exp 1, diarrhea incidence (0-4 dpi) was 10, 51, 49, and 36% for NCC, CC, FBA, and PBA pigs, respectively (NCC vs CC $P < 0.001$; CC vs PBA, $P = 0.089$). BW at dpi 11 was 12.17, 11.86, 11.55, and 12.26kg for NCC, CC, FBA, and PBA pigs, respectively (NCC vs. CC $P = 0.502$; CC vs. FBA, $P = 0.481$; CC vs. PBA, $P = 0.386$; FBA vs. PBA, $P = 0.125$). In Exp 2, ADG was +0.026kg for FBA and +0.032kg for PBA compared with CON (0.263kg; $P \leq 0.001$). ADFI was +0.019kg for FBA and +0.022kg for PBA compared with those fed CON (0.371kg; $P = 0.099$ and $P = 0.055$, respectively). Gain to feed was +0.064 for PBA and +0.035 for FBA compared with CON (0.656; $P = 0.013$ and $P = 0.172$, respectively). Final BW was +0.550kg for FBA and +0.640kg for PBA pigs compared with CON (11.54kg; $P \leq 0.001$). In conclusion, FBA and PBA increased growth performance in nursery pigs, even with pharmacological zinc oxide present in the diet; furthermore, feeding PBA demonstrated additional benefits by decreasing diarrhea and improving feed efficiency of nursery pigs. **Key Words:** Nursery pigs, intestinal modifiers

**79 Effect of medium-chain fatty acid supplementation levels on growth performance, fecal score, and gut permeability in weaning pigs.** M Mejia*[1], J. Torres[1], C. H. Kwon[1], E. Safaie[1], S. Greer[1], E. Davis[2], M. Metz[2], and Y. D. Jang[1], [1]*University of Georgia, Athens, GA, USA*, [2]*Fortiva, Arden Hills, MN, USA*. This study was conducted to evaluate the effects of medium-chain fatty acid (MCFA) supplementation levels on growth performance, fecal score, and gut permeability of weaning pigs. At weaning, a total of 100 pigs [18.4 ± 1.8 d of age; initial body weight (BW) of 5.72 ± 1.4 kg] were allotted to 5 treatments in 5 replicates with 4 pigs per pen for a 35-d feeding trial. Treatments were dietary MCFA (a blend of free-form C6 to C12 fatty acids) supplementation levels of 0.0, 0.2, 0.5, 1.0, and 1.5% for d 0-21 (Phase 1) and a common diet was fed to all pigs for d 21-35 (Phase 2). Growth performance, fecal score (1 = normal, 4 = severe diarrhea), and plasma d-lactate levels, an indicator of gut permeability, were measured. Orthogonal polynomial contrast analysis was performed to evaluate linear and quadratic effects of increasing MCFA levels. With increasing MCFA levels, quadratic increases were observed in BW ($P < 0.05$) at d 7 and 14 (8.50, 8.49, 9.03, 8.79, and 8.69 kg for 0.0, 0.2, 0.5, 1.0 and 1.5% MCFA levels, respectively), while it increased linearly ($P < 0.05$) at d 21, 28, and 35 (19.9, 20.1, 21.2, 21.4, and 21.3 kg, respectively). Average daily gain increased linearly with increasing MCFA levels in d 0-21 and 21-35, while linear and quadratic increases were observed in overall period ($P < 0.05$; 0.428, 0.428, 0.462, 0.472, and 0.467 kg/d, respectively). Average daily feed intake increased linearly ($P < 0.05$) with increasing MCFA levels in d 21-35 and overall period (0.641, 0.640, 0709, 0.693, and 0.695 kg/d, respectively), while gain-to-feed ratio and fecal score were not different in overall period. Plasma d-lactate levels had a quadratic tendency with increasing MCFA levels ($P = 0.10$; 0.296, 0.166, 0.179, 0.232, and 0.348 mM, respectively). In conclusion, increasing MCFA supplementation levels increased overall growth rate and feed intake of nursery pigs from 0.5% supplementation level, while gut permeability was reduced up to 0.5% MCFA levels with no impact on fecal score. **Key Words:** gut permeability, medium chain fatty acid, pigs

**80 EU Circles project: Multi-kingdom microbiota analysis identified distinct microbial networks associated with the supplementation of a blend of chestnut and quebracho extracts, impacting the metabolism of nursery pigs reared under different farming conditions.** P. Trevisi*[1], D. Luise[1], G. Palladino[2], D. Scicchitano[2], G. Babbi[2], A. Castagnetti[3], S. Rampelli[2], M. Candela[2], P. L. Martelli[2], and F Correa[1], [1]*Department of Agricultural and Food Sciences, University of Bologna, 40127 Bologna, Italy*, [2]*Department of Pharmacy and Biotechnology, University of Bologna, 40126 Bologna, Italy*, [3]*Wellmicro srl, 40138 Bologna, Italy*. This study evaluated the effects of tannins from chestnut and quebracho on growth, health, and fecal metagenomic and metabolomic profiles of weaned pigs under two farming conditions. A total of 160 piglets (24 ± 2 days old; T0) were assigned to high (H) or low (L) performance weaning units based on body weight and litter origin. Within each unit, piglets were further divided into 2 dietary groups: i. Control group (CON; 40 pigs) fed a standard weaning diet; ii. Treated group (TRT; 40 pigs) fed the standard diet supplemented with 2 g/kg of the tannin blend. BWs were recorded at days (d) 35 (T1), 49 (T2), and 76 (T3) post-weaning, and fecal samples were collected at each time point for shotgun metagenomic and metabolomic characterization. The data on growth performance and metagenomics were analyzed considering the effects of group (TRT or CTR) and farm (H or L), while metabolites were analyzed using a Sparse Partial Least Squares Discriminant Analysis (sPLS-DA) model. At T2, the diarrhea occurrence was higher in the CON group than in the TRT group (P = 0.02) in farm L. Microbial alpha diversity at T2 and T3 was lower in farm L ($P < 0.05$) and in the TRT group compared to CON ($P < 0.01$). Beta diversity for species and functions was influenced by farm and diet interactions at all time points ($P < 0.05$). At T2, were identified antioxidative metabolites like Urolithin B and Isourolithin A, metabolites derived from microbial degradation of tannins, associated to the TRT group in farm L. Weighted gene correlation network analysis identified *Corynebacterium* and *Pseudomonas* subspecies known for tannase enzyme production (r > 0.5) associated with Urolithin metabolites. In conclusion, tannin supplementation effectively reduced diarrhea occurrence, particularly under poor sanitary conditions, and select a multi-kingdom microbial network able of metabolize tannins and producing antioxidant metabolites able to prevent weaning-associated gut health impairments. **Key Words:** Fecal Metabolome, Tannins, Urolithin

**81 Supplementation of a postbiotic product in gestating and lactating sows positively affect piglets' performances.** B. Polimeni*, F. Correa, D. Luise, A. Zurru, F. Palumbo, M. Benatti, and P. Trevisi, *Department of Agricultural and Food Sciences, University of Bologna, Bologna, Italy*. The genetic selection of hyper-prolific sows has led to challenges like prolonged farrowing, variable piglet birth weights, and higher neonatal mortality due to insufficient intake or quality of colostrum. To improve piglets' survival during suckling, nutritional strategies like postbiotic are considered. The postbiotic, consisting in inanimate microbial cells and/or their component, have been recently introduced in animal husbandry to support gut health and growth performances. This study aims to evaluate the effect of a postbiotic (powder of heat-inactivated Lactobacillus (*L. fermentum and L. delbrueckii*)) supplemented to sows in late gestation (G) and at different doses during lactation (L) on the growth and health of piglets. During the last 3 weeks of gestation, 200 sows were divided into 2 groups fed with a standard gestation diet (G0; 40 sows) or the same diet supplemented with 20 g/t postbiotic (G20; 160 sows). After farrowing, the G20 group was split into 4 groups receiving a different level of postbiotic: 0 (L0), 10 (L10), 20 (L20) or 100 (L100) g/t. Productive performances of the sow were recorded at farrowing; piglets BW and mortality was collected at d0, d6, d14, d28. Data on piglet BW and ADG were analysed using a linear model included the diet, parity and batch as factors and litter size and initial piglet's BW as covariate. The diet influenced piglet BW (d0), with a higher weight of pigs born from G20 sows than G0 ($P < 0.05$). The BW at d6 and ADG from d0 to d6 was higher for the piglets of the sows receiving the postbiotic during gestation ($P < 0.001$). Mortality during suckling was lower in the groups receiving the postbiotic, particularly in G20-L0 and G20-L100, compared to G0-L0 ($P < 0.01$). The supplementation of postbiotics in the gestating diet has been demonstrated to be an effective strategy to improve piglet survival and performance, probably due to a modulation in the quality of colostrum of the sows. **Key Words:** litter performances, sow nutrition, postbiotic

**82 Daily pattern of feeding behavior and SID lysine balance response to low protein diet in weaned pigs.** Yao Zhu* and Jeroen Degroote, *Faculty of Bioscience Engineering, Laboratory for Animal Production and Animal Product Quality, Ghent University, 9000 Ghent, Belgium.* This study investigated daily changes of feeding behaviors, SID lysine requirement, and SID lysine balance (expressed as a ratio of SID lysine requirement and SID lysine intake) under various low protein strategies in weaned piglets housed in electronic feeding stations (EFS). A total of 101 28-day-old piglets (6.90 ± 0.10 kg) were assigned to 10 pens with four treatments for 42 days: low-protein diet (16.2% CP, 10.6 g/kg standardized ileal digestible (SID) lysine) feeding for 10 days (LP10) or 18 days (LP18) followed by starter diet (17.9% CP, 11.8 g/kg SID lysine) until day 42; high-protein diet was fed for 10 days (HP10) or 18 days (HP18) and transitioned to starter diet until day 42. A mixed model with treatment as fixed effect and pen as random effect was used to analyze daily data. Daily visit duration, duration per visit, and interval between visit were not affected by various feeding strategies, while they all showed a stable trend after 15 days. Daily SID lysine intake was significantly lower in LP18 on day 12, 13, 15, 16, and 17 (all $P < 0.05$). Daily gain to feed fluctuated after diet shifts (day11 and 19), but no difference on other days. Daily lysine requirement was significantly lower in LP18 on day 14, 17, and 26 (all $P < 0.05$), while much higher in LP10 on day 17 and 26. SID lysine balance was much higher in HP18 on day 6 ($P < 0.05$) and potentially increasing in HP10 on day 10 ($P < 0.10$). However, cumulative intake, requirement, and balance of SID lysine were significantly fluctuated from day 15 to 22 and day 34 to 38, from day 15 to 19, and from day 14 to 17 and day 25 to 39, respectively. In conclusion, low-protein strategy did not affect feeding behaviors. However, it temporarily impacts daily SID lysine balance or daily gain to feed when shifting feeds, and it may have cumulative effects on piglets over time, which was associated with regular feeding behavior established, specifically after day 15. **Key Words:** feeding behavior, SID lysine balance, low-protein diet

**83 The impact of a novel whey protein concentrate (FXP™) on serum C-reactive protein and intestinal morphology of nursery pigs during a natural enteric health challenge.** S. Rossman*[1], J. Simmons[2], A. Woodward[2], and N. Horn[2], [1]*Iowa State University, Ames, Iowa 50011*, [2]*United Animal Health, Sheridan, Indiana 46069*. Weaning is a stressful event that can exacerbate enteric pathogens and lead to scouring, poor performance, gut lag, and mortality. A novel whey protein concentrate (WPC; FXP™) is hypothesized to improve nursery pig health due to high concentrations of immunoglobins and dairy-derived lipids and proteins, which have been demonstrated to contain anti-pathogenic properties. The objective of this study was to determine the effectiveness of FXP™ on intestinal morphology and inflammatory status of nursery pigs under a postweaning enteric health challenge. At weaning, pigs were allotted based on body weight in a RCDB to 8 replicate pens per treatment with 4 pigs per pen. Pens were assigned to either a control (CTL) or control + 0.60% FXP™ (FXP™) diet for 23 d. A natural, multi-pathogen challenge that included a high prevalence of Rotavirus A, F18 *E. coli*, *S. enterica*, and *S. Suis* presented in all pens throughout the study, resulting in scouring starting at d 10 postweaning and mortality averaging 46% by the end of the experiment. On d 14 and 23, blood was collected from all pigs, centrifuged, and serum was collected, pooled by pen, and analyzed for C-reactive protein (CRP) via ELISA. Furthermore, on d 23, the median pig from each pen was identified, euthanized, and the mid sections of jejunum and ileum were excised. Tissues were subsequently fixed in formalin and slides were made and stained with hematoxylin and eosin for visualization of villi and crypts. On study d 14, FXP™ pigs had reduced *($P < 0.01$)* CRP concentration compared to CTL. In the jejunum and ileum, FXP™ pigs had an increased ($P = 0.05$ and 0.04, respectively) VH:CD compared to CTL due to numerically increased villus height with similar crypt depth measurements in both the jejunum and ileum. These results show FXP™ supplementation can reduce inflammation and improve intestinal health in pigs experiencing a natural enteric health challenge. **Key Words:** nursery pigs, intestinal morphology, health challenge

**84 Effect of pistachio shell powder on growth performance of weanling pigs.** Y Kim*[1], SA Lee[2], and H Stein[1,2], [1]*Division of Nutritional Sciences, University of Illinois at Urbana-Champaign, Urbana, IL, USA*, [2]*Department of Animal Sciences, University of Illinois at Urbana-Champaign, Urbana, IL, USA*. The objective was to test the hypothesis that, despite the high fiber concentration, pistachio shell powder (PSP) does not reduce growth performance of weaning pigs. A total of 160 pigs [initial weight (BW) = 5.23 ± 0.54 kg] were randomly allotted to 4 dietary treatments using a randomized complete block design with initial BW as the block. There were 2 barrows and 2 gilts per pen with10 replicate pens per treatment. The control diet contained mainly corn and soybean meal. Three additional diets were formulated to include 5.0, 7.5, or 10.0% PSP at the expense of corn and metabolizable energy of each diet was 3,400 kcal/kg. More crystalline amino acids and soybean oil were used as PSP increased in the diets to meet the requirements for pigs. Average daily gain (ADG), average daily feed intake (ADFI), and gain to feed (G:F) were calculated for phase 1 (d 1 to 21), phase 2 (d 22 to 42), and for the overall period. Fecal scores were also recorded during phase 1. The statistical model included diet as fixed variable and replicate as random variable. Polynomial contrasts were used to test linear and quadratic effects of increasing PSP in diets. Phase 1 results indicated that ADG and final BW tended to increase as PSP increased in the diet (quadratic; $P = 0.067$). The ADFI significantly increased as PSP increased to 5.0 or 7.5% and decreased as more PSP was used in the diet (quadratic; $P = 0.030$). The G:F tended to increase by increasing PSP in diets (linear; $P = 0.070$). Growth performance during phase 2 and fecal score did not differ among treatments. Overall, ADG and G:F were not significantly affected, but ADFI showed a tendency to increase in a quadratic manner as the inclusion of PSP in the diets increased (quadratic, P = 0.082). In conclusion, with the exception of a tendency for increased ADFI, overall growth performance of weanling pigs was not affected by increasing PSP in diets by 5% that were formulated to meet requirements for energy and digestible amino acids. **Key Words:** corn, pistachio shell powder, weanling pigs

**85 Improved phosphorus digestion and utilization following dietary phosphorus depletion-repletion in growing pigs.** G.C. McKibben*[1], S. Becker[2], P. Wilcock[2], G. Cordero[2], G.A. Gomes[2], and N.K. Gabler[1], [1]*Iowa State University, Ames, Iowa, United States of America*, [2]*AB Vista, Marlborough, Wiltshire, United Kingdom*. Diets depleted in available phosphorus (avP) reduce growth and bone mineralization. Our objective was to evaluate the digestive efficiency and utilization of calcium and phosphorus following avP depletion-repletion feeding. Over a 49-day study, 48 pigs (13.1 ± 0.7 kg BW) were utilized across two experimental periods. In a 35-day period 1 (P1), 8 pens/trt (2 pigs/pen) were fed diets representing adequate (Adeq), marginally deficient (Marg), or deficient (Def) avP formulated to 0.36, 0.17 and 0.08% avP and at a total calcium to avP ratio of 2.91, 3.15 or 6.40, respectively. In the subsequent 14-day period (P2), Marg and Def pigs were fed diets formulated above requirement at 0.54% avP and a 2.19 Ca:avP, while the Adeq pigs remained on the same P1 diet (n=7-8 pens/trt). Period 1 and 2 pig performance, whole body DXA bone mineral content (BMC) and density (BMD), and Ca and P apparent total tract digestibility (ATTD) were measured. Data were analyzed using pen as the experimental unit. Adeq, Marg, and Def BW differed at the end of P1 (40.1, 39.0 and 32.2 kg, respectively, $P < 0.001$) and P2 (53.4, 53.2 and 43.1 kg, respectively, $P < 0.001$). At the end of P2, whole body BMD was greatest in the Adeq compared to Marg and Def (0.93, 0.85 and 0.73 g/cm2, respectively, $P < 0.001$). The Def pigs had the highest ATTD-P coefficient compared to the Adeq and Marg (70.5, 61.8 and 65.4%, respectively, $P = 0.034$) in P2. Nitrogen and organic matter ATTD were higher in the Def and Marg pigs compared to the Adeq pigs ($P < 0.05$). Def Ca-ATTD increased by 18% over the Adeq, with Marg being intermediate ($P = 0.003$). BMC: digestible Ca was higher in Def and Marg pigs compared to Adeq pigs (1.30 and 1.16 vs 0.60 g/g, $P = 0.004$). Expectedly, BMC: digestible P in Def and Marg pigs was higher than Adeq pigs (1.95 and 1.74 vs 0.79 g/g, $P < 0.001$). In conclusion, avP depletion-repletion feeding strategies increased phosphorus and calcium digestibility and utilization in growing pigs. **Key Words:** Calcium, Phosphorus

**86 Supplementation with 2′-Fucosyllactose improve growth performance, jejunal digestion and absorption of nutrients in lactating piglets by increasing the proliferative capacity of stem cells.** W. Wang*[1,2], K. Yu[1,2], and W. Zhu[1,2], [1]*Laboratory of Gastrointestinal Microbiology, Jiangsu Key Laboratory of Gastrointestinal Nutrition and Animal Health, College of Animal Science and Technology, Nanjing Agricultural University, Nanjing 210095, China*, [2]*National Center for International Research on Animal Gut Nutrition, Nanjing Agricultural University, Nanjing 210095, China*. Human milk oligosaccharides (HMOs) have an important impact on the growth, development and health status of newborns. 2′-Fucosyllactose (2′-FL), the most abundant HMOs, which have important nutritional and medical values. To further investigate the effects of 2′-FL on intestinal digestion, absorption and development in neonates, a total of 10 liters of newborn piglets (Duroc × Landrace ×Large White), two piglets were randomly selected from each litter and divided into two groups: control group (CON) and 2′-FL group (2′-FL) with one piglet in each litter for each group (n = 10). The experiment started at 12 hours after the piglets were born and ended at 21 days, all piglets were breast-fed. In addition, piglets in the 2'-FL group were gavaged with 2'-FL at 0.1 g/kg bw per day, whereas piglets of control group were given the same volume of normal saline. The results showed that supplementation with 2'-FL significantly increased final body weight and average daily weight gain of piglets and had higher jejunal intestinal villus height as well as villus to crypt ratio, the mRNA expression levels of glucose, fatty acids, and amino acid transporters and lipase activity ($P < 0.05$). The cell proliferation results indicated that supplementation with 2'-FL promoted the expression of jejunal PCNA at both the mRNA and protein levels ($P < 0.05$). In addition, supplementation with 2'-FL significantly increased the expression level of marker gene in jejunal stem cells ($P < 0.05$). The Wnt signaling pathway, which is important for regulating the proliferation of intestinal stem cells, was significantly increased mRNA expression levels in the jejunum of piglets in the 2'-FL group ($P < 0.05$). In conclusion, supplementation with 2'-FL improved growth performance, jejunal digestion and absorption of nutrients in lactating piglets, possibly due to increased proliferative capacity of stem cells. The underlying mechanisms need to be further explored. **Key Words:** 2'-FL, lactating piglets, stem cell

**87 Supplementation of a consensus bacterial 6-phytase variant increased bone Zn concentration and maintained performance of piglets fed diet without added trace minerals (Zn, Cu, Fe and Mn).** Y Dersjant-Li*[1], G Dusel[2], K Schuh[2], D. E. Velayudhan[1], E. Vinyeta[1], and L Marchal[1], [1]*Danisco Animal Nutrition, IFF, Oegstgeest, The Netherlands*, [2]*University of Applied Sciences Bingen, Berlinstrasse 109, 55411 Bingen am Rhein, Germany*. This study evaluated if a consensus bacterial 6-phytase variant (PhyG) can totally replace added trace minerals (TM; Zn, Cu, Fe and Mn) in piglets. In total 144 DanBred x Pi weaning piglets [28 days of age, initial BW of 7.0 (±0.44) kg] were used in a randomized complete block design with 3 dietary treatments (12 replicates, 4 piglets/pen, 1:1 male/female). Diets were based on corn/SBM and fed in pelleted form in two phases (1-14 and 14-42 days). Treatments comprised: 1) negative control (NC1), nutritionally adequate without added TM; 2) positive control (PC), NC1 supplemented with TM at a commercial level (Zn 120ppm, Cu 80 ppm, Fe 96 ppm, Mn 80 ppm); 3) NC2+PhyG, NC1 reformulated without added inorganic P and reduced Ca, supplemented with PhyG at 1500, 1000 FTU/kg in phase 1 and 2, respectively, without added TM. At day 42, blood and femur bone samples were collected from 1 pig/pen for analysis of TM content. The BW was analyzed individually, feed intake and FCR were analyzed using pen as experimental unit and corrected for mortality. A one-way ANOVA was performed, and treatment means were separated using Tukey's HSD test. The NC1 treatment reduced final BW compared to PC and NC2+PhyG (24.5, 26.5 and 26.3 kg respectively, $P < 0.05$). NC2+PhyG numerically increased ($P > 0.05$) blood Mn (19.6 vs. 15.8 and 14.9 mg/L) and Zn concentration (3.1 vs. 2.5 and 2.5 mg/L) vs. NC1 and PC respectively. PC increased Zn concentration ($P < 0.05$) in bone ash vs. NC1 (211.2 vs 185.5 mg/kg, $P < 0.05$) and NC2+PhyG further increase Zn in bone ash (235.4 mg/kg) vs PC ($P < 0.05$). In conclusion, PhyG phytase improved the bioavailability of Zn from basal ingredients and maintained performance compared to the PC with TM supplementation. The results indicate that PhyG phytase could be used to totally replace added TM especially

Zn ( Fe, Cu and Mn were not affected) at the commercial dose level in piglet diets, which can help to reduce the excretion of these TM and improve production sustainability. **Key Words:** Concensus phytase, bio-availability, trace minerals

**88 The effects of supplementing a blend of polyphenols from chestnut and quebracho extracts with different levels of zinc oxide on the performance and diarrhea incidence in weaned piglets.** V. Cantarelli[1], J.F. Durau[2], G.M. Stingelin[2], J.B. Lancini[3], O. Desrues*[3], N. Panciroli[3], and E. Santin[3], [1]*AnimalNutri Swine Experimental Center, Brazil*, [2]*Labitah Animal Health Ltda., Brazil*, [3]*Silvateam S.p.A., Italy*. The replacement of high level of zinc oxide (ZnO) commonly used in piglet diet to prevent post-weaning diarrhea is a challenge for modern pig production. Plant polyphenols obtained from chestnut and quebracho extracts (PE) have been shown to modulate gut microbiota and contribute to mitigate intestinal disorders in piglets. Here, we evaluated the effects of PE with different doses of ZnO on the growth performance and health status of weaned piglets during 42 days. Four hundred piglets (males/females) weaned at 23 days of age were blocked by initial bodyweight (6.0±0.5 kg) and sex, and were allocated to five treatments (eight replicates per treatment with 10 animals per pen). T1: high level of ZnO (3100 ppm 0-7 d; 2670 ppm 8-14 d; 2220 ppm 15-28 d; 1330 ppm 29-42 d); T2: T1+ PE at 1500 ppm 0-42d; T3:T1+ PE at 3000 ppm (0-14 d) and 1500 ppm (15-42 d); T4: reduced level of ZnO (half dose of T1) + PE at 3000 ppm (0-14 d) and 1500 ppm (15-42 d); T5: PE at 3000 ppm (0-14 d) and 1500 ppm (15-42 d) without ZnO. The analysis of the treatments effects was conducted with a linear mixed-effect model using lmer function and post-hoc Tukey adjustment (R software). The final BW for T1 to T5 were 22.3, 22.1, 21.8, 23.1, 23.0 ± 1.9 kg respectively. The diarrhea incidence from 0-42d for T1 to T5 were 18.8, 12.3, 10.8, 14.7, 22.2 ± 2 % respectively. There was no statistical difference between ZnO (T1) and PE (T5) for final BW and diarrhea incidence. Piglets supplemented with a reduced level of ZnO + PE (T4) tended to be heavier than T3 ($P$ = 0.06). Moreover, the piglets supplemented with ZnO + PE (T2 and T3) had lowest diarrhea incidence from 0-42d, which was statistically significant as compare to T5 ($P$ = 0.01). These results confirmed that a blend of chestnut and quebracho extracts can help preventing diarrhea and increase performance in piglets, and is a suitable solution for holistic approach targeting at the replacement or reduction of ZnO. **Key Words:** polyphenol, piglet, ZnO

**89 Ammonia, methane, and odour in pigs: the role of nutrition in emission pathways.** E.C. Teunissen*[1,2], P. Bikker[1], and A.J.M. Jansman[1], [1]*Wageningen Livestock Research, Wageningen University & Research, Wageningen, The Netherlands*, [2]*Animal Nutrition Group, Department of Animal Sciences, Wageningen University & Research, Wageningen, The Netherlands*. Reducing gaseous emissions from pig production systems is critical for mitigating environmental impact and improving air quality in pig barns. Although the use of by-products in pig diets offers sustainability benefits, their characteristics as part of the diet may also present challenges, such as increased gaseous emissions. This highlights the need for an integrated approach to evaluate the influence of diet composition on gas production, and the role of by-products in sustainability of pig production. This review aimed to define the pathways leading to gaseous production (ammonia, methane, and odour) in pigs and slurry, and identify key nutritional factors affecting these pathways. Ammonia is formed via microbial fermentation of nitrogenous compounds in the large intestine and slurry, and through urea hydrolysis in slurry. Ammonia produced in the large intestine is detoxified by the liver into urea, which is excreted in urine. In the slurry, urea is hydrolysed by microbial urease, releasing ammonia. Methane formation is a two-step process in which microbial fermentation metabolites serve as substrate for methanogenic archaea. Enteric methane is excreted via respiration and flatulence, while methane from slurry arises from fermentation of organic matter present in the slurry, which in turn provides substrates for methanogenic archaea. Odorous compounds result from fermentation in the hindgut and slurry. Some compounds are excreted directly in faeces or urine, while others arise in the slurry from microbial processing of detoxified metabolites or organic matter fermentation. This study emphasizes the importance of optimizing diet composition and feeding strategy to mitigate emissions. By evaluating these pathways and their underlying drivers, it provides a basis for developing nutritional strategies that help minimize the

environmental impact of pig production, while addressing trade-offs in dietary characteristics and use of by-products in pig diets. **Key Words:** emission, pathways, nutrition

**90 Effects of reducing dietary calcium with a fixed STTD P levels on performance, carcass traits and bone characteristics in growing-finishing pigs supplemented with phytase.** C. C. S. Martins*[1], E. Perez-Calvo[2], C. P. Lozano[2], M. F. Prata[1], and C. A. Silva[3], [1]*DSM-Firmenich, São Paulo, SP, Brazil*, [2]*DSM-Firmenich, Kaiseraugst, Switzerland*, [3]*State University of Londrina, Londrina, PR, Brazil*. Excessive dietary calcium (Ca) reduces phosphorus (P) digestibility which may reduce feed intake and growth performance. Thus, nutritional strategies to optimize mineral absorption are required. The study evaluated the effect of reduced 30% calcium (Ca) and fixed available P (STTD) recommended by PIC (higher than NRC 2012) and phytase on performance, carcass traits, blood parameters and bone characteristics in growing finishing pigs. 120 pigs (PIC®, 22.6 kg BW), were allocated in two dietary treatments (High Ca- H and Low Ca-L) with 12 replicates/treatment (5 pigs/replicate). Four feeding phases were offered ad libitum: Growing I (G1 = 28days, H Ca = 0.68% vs L Ca = 0.48% and STTD *P* = 0.34%); Growing II (G2 – 20 days: H Ca = 0.64% vs L Ca = 0.45% and STTD P = 0.33%); Finishing I (F1 – 20 days: H Ca = 0.64% vs L Ca = 0.44% and STTD P = 0.32%), Finishing II (F2 –17 days: H Ca = 0.64% vs L Ca = 0.44% and STTD P = 0.32%). All diets with 600 FTY/kg of phytase and matrix values for Ca of 0.14% and for STTD P of 0.15%. Average daily gain (ADG), feed intake (ADFI), and feed conversion ratio (FCR) were calculated by phase. Blood samples were collected from same one pig/replicate at the start and the end of each feed phase. At the end of the trial, the third metacarpal was collected to analyze the mineral content and bone breaking strength (BBS). All data were analyzed using SAS software to compare the means by Tukey test ($P < 0.05$). L Ca increased ($P < 0.05$) live weights at G1, G2, and F1 and improved ($P < 0.05$) ADG and FCR only at G1. L Ca pigs also increased serum P levels ($P < 0.05$) and carcass yield ($P < 0.05$). Bone ash and P content were also increased ($P < 0.05$) in L Ca, whereas Ca content and BBS were similar. However, no differences were detected on ADG, ADFI and FCR at overall period between treatments. In conclusion, reducing Ca levels by 30% with fixed STTD P and using matrix values for phytase optimization, improves P absorption, carcass yield, and bone characteristics. **Key Words:** mineral nutrition, digestibility, performance

**91 Effects of supplementing pure vitamin E vs lower levels of vitamin E in combination with a polyphenol compound on growth performance and oxidative stress in weanling pigs.** L. Blavi*, L. Sobrevia, S. Laird, M. A. Ton Nu, S. Tibble, and A. Koppenol, *AB Neo, Fraga, Huesca, Spain*. Vitamin E (Vit E) is a powerful antioxidant but an expensive ingredient. The blend of polyphenols (PP) with lower Vit E has shown similar effects. The trial aimed to compare the supplementation of only Vit E vs the combination of a PP compound with lower levels of Vit E on growth performance and oxidative status in post-weaned pigs. A total of 1953 pigs (24d old, 6.51 ± 0.89 kg body weight (BW)) were distributed into 2 treatments (20-21 pigs/pen, n=48) with 150 ppm of Vit E (VE) or 75 ppm equivalents of Vit E (45 ppm Vit E + 6g PP (extract of grape and olive), VEP) for 28d (14d phase 1 and 14d phase 2), then all pigs were fed a common diet for 14d (phase 3). Growth performance, mortality, and culling was measured by phase. On d 9, 23, and 37, blood samples were collected (n=48) for total antioxidant capacity (TAC), total oxidant status (TOS) and Haptoglobin (Hp). Data were analyzed with fixed effect of treatment and initial BW as covariate. Pigs fed VEP compared to VE had greater ($P < 0.001$) BW on all phases (d 42: 19.78 vs 19.05kg), greater ($P < 0.001$) daily gain on phase 1, 2, and globally (321.5 vs 303.5 g/d), and better FCR in phase 1, 3, and globally (1.262 vs 1.303). No differences ($P > 0.1$) were observed on daily feed intake, mortality, and culling. Pigs fed VEP had lower ($P < 0.05$) Hp (0.03 vs 0.13g/L) in phase 1 but tended ($P = 0.066$) to have higher Hp in phase 2 (0.55 vs 0.35g/L). No differences ($P > 0.1$) were observed on TAC and TOS, except on phase 3, where pigs fed VEP had lower ($P < 0.05$) TOS levels than pigs fed VE (15.72 vs 22.2 µmol/l). Pigs fed only Vit E had higher TOS:TAC values than pigs fed the combination of vit E and PP. To conclude, using lower levels of Vit E in combination with PP improved growth performance, reduced

inflammation at the beginning of the post-weaning period and improved the prooxidant-antioxidant balance. The results suggested further research on the synergistic effects of combining Vit E and polyphenols. **Key Words:** Vitamine E, Polyphenols, oxidative stress

**92 Effect of increasing insoluble dietary fiber levels on growth performance and microbiota in weanling pigs.** L. Blavi*, L. Sobrevia, S. Laird, M. A. Ton Nu, S. Tibble, and A. Koppenol, *AB Neo, Fraga, Huesca, Spain*. Fiber specifications are becoming more common in diet formulation after weaning despite no recommendations being published. The objective was to evaluate the effect of increasing insoluble dietary fiber (iDF) levels on performance and fecal microbiota composition in post-weaning pigs. A total of 2131 pigs (24 d old, 5.76 ± 0.97 kg BW) were distributed into 5 treatments (24 pigs/pen, n = 18) with increasing iDF levels at 6, 9, 12, 15 and 18% in phase 1 (d0-14) and 8, 11, 14, 17, and 20% in phase 2 (d14-28) and a common diet with 16% iDF in phase 3 (d28-42). Growth performance, mortality, and culling was measured by phase. On d42, pooled fecal samples were collected (n=8). Data were analyzed using linear and quadratic orthogonal polynomial contrasts for equally spaced treatments. Increasing iDF linearly decreased body weight, gain, feed intake, and increased feed conversion ratio ($P < 0.05$) in all phases, except feed intake in phase 3. The highest growth was for 9% and 11% iDF in phase 1 and 2, respectively. No effect on mortality and culling, except in phase 3 where pigs fed the highest iDF levels had higher mortality (0.24 vs 0.00%) than the rest. Overall pigs fed 9 and 12% iDF in phase 1 and 11 and 14% iDF in phase 2 had numerically the lowest mortality, culling, and medication. Formulating with 18% and 20% iDF in phase 1 and 2, respectively, significantly increased ($P < 0.05$) the main butyrate-producing bacteria families (*Lacnospiraceae* and *Ruminococcaceae*) and tended to increase *Bifidobacterium* ($P = 0.053$). However, there was no effect of iDF on total short chain fatty acids (SCFA) or its profile ($P > 0.05$). The optimal levels of iDF for growth and health were achieved at 9 and 11% iDF in phase 1 and 2, respectively. At d42, 2 weeks post common dietary phase, there were no significant differences among treatments on microbiota composition and on SCFA, which suggest that earlier collections would have been more interesting to analyze. **Key Words:** Insoluble fiber, growth performance, microbiota

**93 Self-supplementation of amino acids by piglets under different sanitary conditions in a choice-feeding setting.** I. Minussi*[1], A. J. M. Jansman[2], J. E. Bolhuis[1], and W. J. J. Gerrits[1], [1]*Wageningen University & Research, Wageningen, The Netherlands*, [2]*Wageningen UR, Livestock Research, Wageningen, The Netherlands*. Low sanitary conditions (LSC) resulting from poor hygiene alter amino acid (AA) metabolism of pigs, changing their AA requirements. Pigs can select a diet that meets their AA requirements over an AA-deficient one when given the choice, suggesting potential for self-supplementation of AA in conditions affecting AA requirements. This study investigated whether self-supplementation for provision of an AA deficient diet in a choice-feeding setting differs for piglets kept under high (HSC) or low sanitary conditions. In a 2×2 factorial experiment, 60 weaned female piglets (TN70 × Tempo) were housed in pairs under HSC or LSC and offered either a diet deficient in eight AA ($LP^-$) (No choice) or the choice between the $LP^-$ and a diet enriched with eight AA ($LP^+$) (Choice) for 19 days. Average daily feed intake (ADFI), gain (ADG), and blood immune parameters were recorded, and analyzed with linear mixed models with fixed effects of sanitary conditions, choice, their interaction, and batch (with period included for performance), and a random effect of pen. LSC piglets had 31, 44, and 21% lower ADFI, ADG, and G:F, respectively, than HSC piglets ($P < 0.01$), while no effect of diet choice was observed. Both LSC and HSC piglets ingested more $LP^-$ than $LP^+$ diet ($P < 0.001$). Over time, percentage of $LP^+$ consumed out of the total feed intake doubled (from 6.5% to 13.2%) for HSC piglets, but decreased (from 10.6% to 1.4%) for LSC piglets ($P < 0.01$ for SC × Period). LSC piglets had 22 and 38% higher blood leukocyte and monocyte counts ($P < 0.01$ for both), respectively, and a 129% higher serum haptoglobin ($P < 0.05$) than HSC piglets. Neutrophil (+53%) and eosinophil (+43%) counts were higher in LSC pigs (both $P < 0.001$). LSC, while decreasing performance and activating the immune system, did not increase the preference for an AA enriched diet. Thus,

piglets kept under LSC choose not to compensate for a higher usage of AA by the immune system, showing reduced priority for growth. **Key Words:** Amino acid, Choice-feeding, Hygienic conditions

**94 Choice white grease equivalence of fat emulsifier in diets fed to growing pigs.** SA Lee*[1], V Perez[2], and H Stein[1], [1]*Department of Animal Sciences, University of Illinois at Urbana-Champaign, Urbana, IL, USA*, [2]*Kemin Ind., Des Moines, IA, USA*. The objective was to test the hypothesis that increasing choice white grease (CWG) and adding a fat emulsifier to diets increase growth performance of pigs. The second objective was to determine the CWG equivalence of the fat emulsifier. Four diets contained corn, soybean meal, and distillers dried grains with solubles and 4 levels of CWG (i.e., 0, 2, 4, or 6%). Two additional diets contained 0 or 2% CWG and a fat emulsifier. A total of 192 pigs (initial weight = 29.9 ± 2.4 kg) were fed ad libitum for 28 days and allotted to the 6 diets with 2 pigs per pen ($n$ = 16). Average daily gain (ADG), average daily feed intake (ADFI), and gain to feed (G:F) were calculated. Data were analyzed using the Proc Mixed of SAS. The statistical model included diet as fixed variable. Linear and quadratic effects of increasing CWG were tested and linear regression equations were developed. The CWG equivalence was then back-calculated using G:F of pigs fed the fat emulsifier diets. Overall, increasing CWG in diets increased (linear; $P < 0.05$) ADG and G:F of pigs, but ADFI was not affected. Supplementation of the emulsifier increased ($P = 0.026$) ADG of pigs without affecting ADFI or G:F. Regression analysis indicated that G:F increased ($P < 0.05$) by 0.00678 for each percentage unit change of CWG inclusion in diet, and G:F of pigs fed the fat emulsifier diets was similar to the G:F of pigs fed diets containing -1.1% or 6.8% CWG for the diets containing 0 and 2% CWG, respectively. In conclusion, growth performance of pigs was improved by CWG and fat emulsifier. The effects of the fat emulsifier were close to 0 when no fat was used, which indicated that the presence of fat with saturated fatty acids is needed to obtain a response to the emulsifier. Although outside the range, G:F of pigs fed the 2% CWG diet with the emulsifier was equivalent to G:F of pigs fed 6.8% CWG diet, which suggests that the emulsifier may contain the energy equivalent to 4.8% CWG (i.e., 343 kcal of net energy). **Key Words:** emulsifier, equivalence, fat

**95 Effects of a Novel Whey Protein Concentrate (FXP™) on Adhesion of Enterotoxigenic *Escherichia coli* F4 and F18 in Intestinal Epithelial Cells.** Hang Lu*[1], Julie Simmons[1], Karely Cantu[1], Nathan Horn[1], Adrienne Woodward[1], Joel Spencer[1], and Aaron Gaines[2], [1]*United Animal Health, Sheridan, IN, USA*, [2]*Ani-Tek, Social Circle, GA, USA*. Enterotoxigenic *Escherichia coli* (ETEC) F4 and F18 are common Gram-negative extracellular pathogens and a major causative agent of post-weaning diarrhea and intestinal dysfunction in nursery pigs. The novel whey protein concentrate (WPC; FXP™) is hypothesized to improve nursery pig performance due to high concentration of immunoglobins and dairy-derived lipids and proteins which have been demonstrated to contain anti-pathogenic properties. The objective of the current research was to determine the effects of FXP™ on the adhesion of ETEC F4 or F18 in IPEC-J2 or IPEC-1 cell lines, respectively, in a series of two experiments. Experimental methodology was similar in each experiment. Prior to the adhesion experiment, ETEC were cultured in Tryptic Soy Broth at 37°C overnight and washed with PBS 2 times before resuspended in serum free culture media (SFM) to obtain the desired multiplicity of infection of 10:1. Mature IPEC cells were treated with either SFM or 0.5% FXP™ dissolved in SFM for 1 hour before exposed to ETEC for 30 min. Cytotoxicity was measured by lactate dehydrogenase release and remained below 20% for all experiments. Cells were then washed and lysed with 0.1% Triton-X for plating on MacConkey Agar and incubated for 24 hours at 37°C prior to colony enumeration. In both experiments, 9 replicates were performed for each treatment and data were analyzed using SAS GLM procedure. An Average of 6.11 $\log_{10}$CFU/mL of F4 (out of 8.89 $\log_{10}$CFU/ml added bacteria) were adherent to IPEC-J2 cells while about 3.89 $\log_{10}$CFU/mL of F18 (out of 8.23 $\log_{10}$CFU/mL added bacteria) were adherent to IPEC-1 cells. FXP™ reduced F4 adhesion in IPEC-J2 cells (5.73 vs 6.49 $\log_{10}$CFU/mL, $P < 0.01$) and tended to reduce F18 adhesion to IPEC-1 cells (3.36 vs 3.93 $\log_{10}$CFU/mL, $P = 0.07$). In conclusion, the novel WPC (FXP™) effectively reduced the adhesion of ETEC F4 and F18 to the epithelial cells *in vitro*

making it a good candidate for growth and health improvement in weaned pigs. **Key Words:** Enterotoxigenic *Escherichia coli* Whey Protein Concentrate IPEC-1

**96 Effects of Organic Acid Supplementation via Water on the Performance of Weaned Piglets.** K.V.Z. Augusto*[1], G. Heim[2], B. Pellicci[3], and A.M. Silvestrim[1], [1]*Trouw Nutrition, Campinas, São Paulo, Brazil*, [2]*Trouw Nutrition, Ameersfort, The Netherlands*, [3]*CEAPA, São Manuel, São Paulo, Brazil*. The use of organic acids (OA) in animal nutrition has been studied due to their potential to support intestinal health and zootechnical performance. These benefits are associated with the reduction of gastric pH, which enhances enzymatic activity, particularly pepsin, and a balanced intestinal microbiota. This study evaluated the effects of a synergistic blend of free and buffered OA applied via water (SWA) on performance and fecal consistency of weaned piglets. The study was conducted at CEAPA, Brazil, using 240 26 d weaned piglets (TN70) (1:1 male-to-female), weighing in average 7.7 ± 0.2 kg. The experimental design was completely randomized with 2 treatments (12 pen/treatment, 10 piglets/pen: Control (corn- and soy-based basal diet) and SWA (same basal diet plus 1 L/1000 L water of SWA during the first 37 d post-weaning). Animals had *ad libitum* access to water and 3 phases feed (Pre-starter; Starter I and II). Evaluated parameters included BW, ADG, ADFI, FCR. Feces were scored daily: normal (1), loose (2), and liquid (3). Data were analyzed with variance analysis (ANOVA, SAS) and statistically with 5% significance level. Final BW (+480 g; +2.2%) and FCR (-3.7 points; -2.4%) were numerically improved by SWA compared to the control group ($P > 0.05$). There was no treatment effect on ADFI and ADG. There was a numerical reduced proportion of piglets with liquid feces during the pre-starter phase in the SWA groups compared to the control (32.6% vs. 43.2%, $P > 0.05$). No relevant differences were observed in subsequent feeding phases. The supplementation of SWA showed potential to improve performance and reduce the incidence of liquid feces during the initial phase of the study. Although the differences were not statistically significant, the results suggest benefits related to intestinal health maintenance and support for zootechnical performance. Further studies are recommended to confirm and expand on these findings. **Key Words:** Feed Additives, water, acidifying

**97 Effect of dietary Acid-Binding Capacity (ABC) and Crude Protein (CP) level on post-weaning pig growth and health.** J.P. Glynn*[1,2], G.E. Gardiner[1], and P.G. Lawlor[2], [1]*Department of Science, South East Technological University, Waterford, Ireland*, [2]*Pig Development Department, Animal & Grassland Research & Innovation Centre, Teagasc, Moorepark, Fermoy, Co.Cork, Ireland*. Reducing dietary acid-binding capacity (ABC) and crude protein (CP) has been proposed to improve post-weaning (PW) pig growth and health. The objective was to determine the effect of reducing dietary ABC and CP for 9 days PW on pig growth and health. At weaning (29 ± 0.95 days), 96 pen pairs of the same sex and of similar weight (8.2 ± 1.1 kg) were randomly assigned to treatment as follows: 1. 21% CP and high ABC-4 (504 meq/kg); 2. 21% CP and medium ABC-4 (440 meq/kg); 3. 21% CP and low ABC-4 (376 meq/kg); 4. 17% CP and high ABC-4 (407 meq/kg); 5. 17% CP and medium ABC-4 (342 meq/kg); 6. 17% CP; low ABC-4 (278 meq/kg). The experiment was a 2x3 factorial arrangement with factors being CP [21% (high) and 17% (low)] and ABC-4 (high, medium, low). High, medium and low ABC-4 levels were obtained by including 10, 5 and 0g/kg limestone flour, respectively, in the diet. Experimental diets were fed for 9 days PW after which common link (D9-20 PW) and grower (D20-35 PW) diets were fed. Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated and analyzed using PROC MIXED in SAS. Diarrhea prevalence was recorded from D1-14 PW and analyzed using PROC GLIMMIX. There was a CP by ABC interaction for diarrhea prevalence ($P = 0.01$). Diarrhea prevalence was reduced when ABC was low compared to high on the low CP diet while this was not the case with the high CP diet. There was a CP by ABC interaction for FCR from D0-9 PW ($P < 0.01$) with FCR on the high ABC and high CP diet being better than that for all ABC levels on the low CP diet but not for other ABC levels on the high CP diet. From D0-9 PW, ADFI was higher on the low than on the high CP diet ($P < 0.01$). In conclusion, reducing dietary ABC reduced diarrhea prevalence when a low but not a high CP diet was fed. Feeding a high CP diet reduced

ADFI and improved FCR from D0-9 PW compared to a low CP diet. **Key Words:** Acid-binding capacity, crude protein, post-weaning pig.

**98 Wheat bran and *Palmaria palmata* as functional ingredients for post-weaning piglets.** Élisabeth Chassé*, Mihai-Victor Curtasu, and Knud Erik Bach Knudsen, *Aarhus University, Viborg, Denmark*. The aim of this study was to evaluate the potential of seaweed *Palmaria palmata* as a functional feed for post-weaning piglets. The hypothesis was that increased levels of soluble fibers as mixed β(1-4, 1-3) xylans from *P. palmata* would be rapidly fermented and could promote gut health. Piglets (6.21 ±1.15 kg BW, n = 32) were weaned at 28 d. Piglets were housed in pairs from a same sow in each pen. The experiment had 2 blocks of 16 piglets each fed the diets for 21 d *ad libitum*. The diets were rice and fish meal based so that the main source of fibers would originate from *P. palmata* (PP) or wheat bran (WB). Fiber content was 6.6% of total NSP and 2.5% of soluble NSP for PP and 6.0% of total NSP and 0.9% soluble NSP for WB. Plasma samples were collected from piglets at d21 and after they were euthanized and necropsied. Statistical model included diet as main effect and pen and sow as random effects (SAS software). Plasma metabolomics data was normalized using pareto scaling and analyzed with an ANOVA with Fisher's LSD *post-hoc* test. Piglet fed PP tended to have lower average daily gain (245 g) compared to piglets fed WB (287 g) for the 0 to 21 d period ($P = 0.052$). Feces dry matter was increased in WB by 18.9% at d7 ($P = 0.006$) and 24.5% at d21 ($P < 0.001$) compared to PP. Changes in plasma metabolome were observed in the direction of WB where lysophospholipids (LysoPC 18:0, LysoPC 18:2, LysoPC 18:1, LysoPC 16:0), 5-aminovaleric acid betaine and hippuric acid were increased compared to PP ($P < 0.05$). Plasma metabolites indicated a diet-origin and were mainly related to intake of grains. Thyroid gland of piglets fed PP tended to be 7.7% larger than WB ($P = 0.075$) which could be explained by a higher content in iodine in the PP diet. In conclusion, inclusion of *P. palmata* to piglet diets as a functional feed did not show improvement of health parameters and special regard on inclusion level should be placed on the impact of toxic minerals when feeding seaweeds to piglets. **Key Words:** Seaweed, fibre, gut health

**99 Spray-dried plasma as a functional protein in weaned pig diets with or without mycotoxins.** L. K. F. Müller[1], A. S. da Silva[1], D. Paiano[1], L. F. S. Rangel[2], J. D. Crenshaw*[2], and J. Polo[2], [1]*Universidade de Estado de Santa Catarina, Chapecó, Santa Catarina, Brasil*, [2]*APC LLC, Ankeny, Iowa, USA*. A study assessed the use of spray-dried plasma (SDP) as a functional protein in weaned pig diets with or without mycotoxins (MYC). Male pigs (7.4±0.4 kg) weaned at 26 days were used in a 2 x 2 factorial design (with/without SDP and with/without MYC, 8 ppm fumonisin + 300 ppb aflatoxins). SDP was included in diets at 6% and 4% during phase 1 (1-7 days) and phase 2 (8-14 days), respectively, and excluded during phase 3 (15-35 days), with diets within phase otherwise kept similar except for MYC levels. There were 6 replicates per treatment with 2 pigs per pen. Pig weight and feed used were recorded at the end of each phase (7, 14, and 35 days) to calculate average daily gain (ADG), feed intake (FI), and feed conversion ratio (FC). Blood samples (5 mL via cranial vena) were collected on weighing days to measure serum biochemical indicators (Albumin - ALB, Cholesterol, and Glucose) and liver health indicators (Cholinesterase - CHE, Alkaline Phosphatase - ALP, Aspartate Aminotransferase - AST, and Alanine Aminotransferase - ALT). Data were analyzed using a 2 x 2 factorial design, with significant interactions prompting further analysis. Results showed no interaction (SDP*MYC) for zootechnical performance variables, with higher ADG and FI for pigs fed diets with SDP at all evaluated phases. ADG and FI was higher in the first week for pigs fed diets with MYC but considering the total period (0-35 days), FI and ADG were lower in the treatments with mycotoxins. Serum ALB levels were higher in SDP treatments on day 35, with no effects on other biochemical serum variables. SDP reduced AST at day 14, while CHE, AST, and ALT levels were higher in MYC diets on day 35, suggesting a cumulative MYC effect. ALP was consistently higher in pigs with MYC diets. Overall, MYC diets increased liver injury markers, lowering ADG and FI. Meanwhile, SDP treatments reduced ALT at the end of the second week and improved feed intake and weight gain in pigs. **Key Words:** pigs,

spray-dried plasma, mycotoxins

**100 Effect of supplementing a *Bacillus subtilis* based probiotic on the concentration of free amino acids in serum of heat stressed growing pigs.** A. Morales*[1], M. Cervantes[1], F. González[1], J.A. Valle[1], S.M. Mendoza[2], and J.K. Htoo[3], [1]*ICA-Universidad Autónoma de Baja California, Mexicali, B.C., México*, [2]*Evonik Corporation, Kennesaw, GA, USA*, [3]*Evonik Operations GmbH, Hanau-Wolfgang, Germany*. Exposing pigs to high ambient temperature (AT) causing heat stress (HS) results in damage to the small intestine epithelia, likely affecting its absorption capacity. Probiotics, in contrast, may help preventing damage to intestinal epithelia. A trial aimed to analyze the effect of a probiotic on serum concentration (SC) of free amino acids (AA) in HS pigs during the absorptive phase peak, was conducted with 12 pigs (21±2 kg BW) in a completely randomized design (4 replicates/treatment. Treatments were: TN-C, thermal neutral pigs (AT, 22 - 24°C) fed a control wheat-soybean meal diet (CP 12.7%, SID Lys 0.94%); HS-C, HS pigs (AT: 26.4 - 35.6°C) fed control diet; HS-P, HS pigs with control diet added with a *Bacillus subtilis*-based probiotic ($1\times10^6$ CFU/g feed). Pigs were fed *ad libitum* during the 21-d trial. On day 21, after a 12-hour fast, pigs consumed 1.0 kg feed in full in less than 15 min. At 2 hours after pigs finished their meal, blood samples were collected from the jugular vein; serum was separated to analyze the SC of free AA. The SC of essential AA (µg/dL) for TN-C, HS-C, and HS-P were: Arg 38.5, 29.3, 34.9; His 15.8, 10.4, 10.5; Ile 15.1, 16.4, 15.3; Leu 25.2, 21.8, 22.0; Lys 42.5, 14.8, 24.2; Met 8.3, 5.1, 4.9; Phe 17.4, 9.3, 11.5; Thr 28.7, 13.9, 15.6; Trp 19.5, 10.1, 9.9; Val 36.2, 31.4, 30.6, respectively. The SC of essential AA, except for Ile, was lower in HS-C pigs, than in TN-C pigs ($P < 0.05$). Serum Lys was higher in HS-P pigs than in HS-C ($P < 0.01$), probably associated with higher abundance of Lys transporter in duodenum. The SC of other essential AA did not differ between HS-C and HS-P pigs. Regarding non-essential AA, the SC of Asn, Asp, Pro, Ser, Tyr decreased in HS-C, compared to TN-C pigs ($P < 0.05$) but it did not differ between HS-C and HS-P pigs. In conclusion, these data confirm the negative impact of HS on absorption of AA, but also indicate that supplemental probiotics in the diet of HS pigs have limited effect on the SC of free AA. **Key Words:** Pigs, probiotic, amino acids.

**101 Efficacy of a Novel Whey Protein Concentrate (FXP™) in Reducing *Salmonella* Adhesion and Invasion in Porcine Intestinal Epithelial Cells.** N Horn*[1], A Woodward[1], J Spencer[1], A Bhunia[2], and A Gaines[3], [1]*United Animal Health, Inc., Sheridan, IN, USA*, [2]*Department of Food Science, Purdue University, West Lafayette, IN, USA*, [3]*Ani-Tek, Social Circle, GA, USA*. *Salmonella* are common gram-negative intracellular pathogens and are a major causative agent of post-weaning diarrhea, mortality, morbidity, and intestinal dysfunction in nursery pigs. The novel whey protein concentrate (WPC; FXP™) is hypothesized to improve nursery pig performance and health due to high concentration of immunoglobins and dairy-derived lipids and proteins which have been demonstrated to contain anti-pathogenic properties. The aim of the study was to determine the efficacy of FXP™ to mitigate adhesion and invasion of *Salmonella choleraesuis, enteritidis,* and *typhimurium* species in porcine intestinal epithelial cells (IPEC-J2). Each *Salmonella* species was evaluated in a separate experiment and there was a minimum of 4 replicates per treatment. Treatments consisted of 1.) control (no pathogen, no FXP™), 2.) pathogen only, and 3.) pathogen + 0.50% FXP™. Mature IPEC-J2 cells were exposed to FXP™ alone for 1 h and *Salmonella* was added to the media for 1 h at a multiplicity of infection of 50:1. Cytotoxicity was measured by lactate dehydrogenase release and remained below 20% for all experiments. Following the 2 h experiments adherent and invaded *Salmonella* were plated on Xylose Lysine Deoxycholate agar for enumeration. Further, cellular integrity was determined by FITC-dextran passage. Co-incubation of FXP™ reduced adhesion of all three *Salmonella* species to IPEC-J2 cells ($P < 0.05$) and reduced invasion ($P < 0.05$) of *choleraesuis* and *enteritidis*, by 54 and 90%, respectively. Exposure of all three *Salmonella* species to cells tended to increase FITC-dextran passage which was partially mitigated by FXP™ ($P < 0.10$). Results from the current experiment show that FXP™, a novel WPC technology, has anti-pathogenic properties against *Salmonella* species and can partially mitigate effects on porcine epithelial cell integrity. **Key Words:** *Salmonella*, Whey Protein Concentrate, IPEC-J2

**102 Protective Effects of a Novel Whey Protein Concentrate (FXP™) on Porcine Rotavirus-Induced Epithelial Damage.** N Horn*[1], A Woodward[1], J Spencer[1], A Bhunia[2], and A Gaines[3], [1]*United Animal Health, Inc., Sheridan, IN, USA*, [2]*Department of Food Science, Purdue University, West Lafayette, IN, USA*, [3]*Ani-Tek, Social Circle, GA, USA*. Porcine rotavirus is a double-stranded RNA virus that infects intestinal epithelial tissue of neonatal and weaned pigs and is a major causative agent of intestinal dysfunction leading to scouring, morbidity, and mortality. The novel whey protein concentrate (WPC; FXP™) is hypothesized to improve nursery pig performance due to high concentration of immunoglobins and dairy-derived lipids and proteins which have been demonstrated to contain anti-viral properties. The aim of the current experiment was to determine the efficacy of FXP™ to mitigate infectivity of porcine rotavirus using a porcine epithelial cell culture model. Porcine rotavirus (serotype A) was isolated from scouring commercial nursery pigs, and MA-104 and IPEC-J2 cells were used as the in-vitro propagation and infectivity cell lines, respectively. There was a minimum of 5 replicates per treatment and treatments consistent of 1) Control (no pathogen, no FXP™), 2) rotavirus only, and 3) rotavirus + 0.50% FXP™. Cytotoxicity was measured by lactate dehydrogenase release and infectivity was measured via immunofluorescence. Further, epithelial integrity was assessed through measurement of trans-epithelial electrical resistance (TEER) and FITC-dextran passage. Compared to control cells, exposure of porcine rotavirus to IPEC-J2 cells resulted in 26% cytotoxicity, virus infectivity of 11 fluorescent focal units (FFU) per viewing field, a reduction in TEER and an increase in FITC-dextran passage ($P < 0.05$). Cells incubated with rotavirus + FXP™ showed a reduction in cytotoxicity (10%) and infectivity (3 FFU/viewing field) ($P < 0.01$) along with a tendency for increased TEER and reduced FITC-dextran passage ($P < 0.10$) when compared to cells exposed to rotavirus only. Results from the current experiment show that FXP™, a novel WPC technology, has anti-pathogenic properties that can partially mitigate infectivity of porcine rotavirus and reduce viral effects on epithelial cell damage. **Key Words:** Rotavirus, Whey protein concentrate, IPEC-J2

**103 Chlorhexidine nanoparticles as alternatives growth promoters show beneficial effects on digestibility of weaned piglets.** A.C.R. Oliveira[1], A.L.B. Mezzina[1], N.A.C. Gomes[1], F.A. Coelho[1], C. Veloso[1], J.A.E. Martínez[1], F.S.S. Tavares[1], F.M. Santos[1], H. Silveira[2], and C.A.P. Garbossa*[1], [1]*University of São Paulo, Pirassununga, São Paulo, Brazil*, [2]*Brazilian Nano Feed, Santo André, São Paulo, Brazil*. This study evaluated the effects of chlorhexidine nanoparticles on nutrient digestibility in nursery piglets. A 42-day experiment was conducted with 96 piglets (barrows and gilts) weaned at 21 days of age, allocated to four treatments in a randomized block design based on initial weight and sex: CON, C25, C50, and C75 (0, 25, 50, and 75 mg/kg of nano-chlorhexidine, respectively), with eight replications per treatment. Each pen of three piglets was considered an experimental unit. Three days before fecal collection, piglets were fed treatment-specific diets containing 0.5% titanium dioxide (TiO2) as a digestibility marker. On day 42, pooled fecal samples (150 g per pen) and feed samples were analyzed for bromatological composition to calculate apparent digestibility of dry matter, mineral matter, crude protein, ether extract, calcium, and phosphorus. Data were analyzed using ANOVA and regression in SAS (version 9.4). A linear decrease in ether extract digestibility ($P = 0.01$) was observed as chlorhexidine nanoparticle inclusion increased, with a reduction of approximately 2.1% across the inclusion range. Quadratic effects were noted for dry matter ($P = 0.003$), crude protein ($P < 0.0001$), mineral matter ($P = 0.0004$), and calcium ($P = 0.004$), with optimized inclusion levels of 34.37 mg/kg, 30.71 mg/kg, 35.73 mg/kg, and 45.11 mg/kg, respectively. These findings indicate that, at optimal levels, nano-chlorhexidine improved nutrient digestibility compared to the control group. In conclusion, chlorhexidine nanoparticles optimized at specific levels enhanced nutrient digestibility of key nutrients and represent a potential alternative to antimicrobial growth promoters in piglet diets. **Key Words:** Nanotechnology, swine, nutrition.

**104 Boosting weight uniformity in nursery pigs with Bacillus-based probiotics: A meta-analysis.** L. H. B. Hansen*, L. Raff, and J. N. Jørgensen, *Novonesis, Animal Biosolutions, Kongens Lyngby, Denmark*. Modern swine production aims to maximize productivity, but weight uniformity (WU) remains a challenge, impacting feed efficiency, labor, and management from early life to slaughter. To our knowledge, no studies have evaluated the effect of probiotic supplementation during nursery on WU. This study evaluated the effect of *Bacillus*-based probiotics on WU in nursery pigs and identified the body weight (BW) changes driving this impact. The meta-analysis included 8 studies across various geographical locations. Pigs were fed nursery diets supplemented with *B. subtilis*–541 and *B. amyloliquefaciens*–516 as single strains or in combination for 42 days (n = 1056) and compared with pigs not supplemented with probiotics (n=1053). Individual BW was recorded at the beginning and end of the nursery phase. Data from the 8 studies were pooled after confirming similar BW distribution patterns. The shift function analysis was used to compare BW distribution across deciles, and logistic regression models to assess shifts in BW groups (R software). Initial BW and WU were identical for both groups (7.2±1.4 kg and 5.5±1.05 CV%). There was no significant difference in mean BW between groups at study end (22.2±4.7 vs. 22.6±4.3 kg). Probiotics improved WU at the end of the nursery phase by reducing CV (12.1±2.0 vs 10.3±2.0 CV%, $P < 0.001$). Pigs from the probiotic group showed less spread BW distribution specifically in the extreme BW groups, and especially in lower BW groups (-0.78 deviation, $P < 0.02$). The uniformity improvement achieved with probiotic supplementation was driven by an increase in BW of smaller pigs (12% vs. 18.6% shifting from low to high deciles, $P < 0.02$). A higher proportion of probiotic administered pigs tended to shift from medium to high deciles (6.4% difference, $P < 0.10$), while a lower proportion moved from medium to low deciles (5.4% difference, $P < 0.10$). In conclusion, probiotic supplementation during the nursery phase reduced BW variability, particularly in smaller pigs. **Key Words:** Probiotics, Weight uniformity, gut health

**105 Supplementation with guanidinoacetic acid improves growth performance and protein deposition of heat-stressed growing pigs.** L. D. Campos*[1], D. A. Marçal[1], L. Hauschild[1], B. Jayaraman[2], and J. K. Htoo[3], [1]*Department of Animal Science, São Paulo State University (UNESP), School of Agricultural and Veterinary Sciences, Jaboticabal, São Paulo, Brazil*, [2]*Evonik Methionine (SEA) Pte. Ltd., Singapore*, [3]*Evonik Operations GmbH, Hanau-Wolfgang, Hesse, Germany*. Guanidinoacetic acid (GAA) is a natural amino acid derivative of arginine and glycine and the only immediate precursor of creatine, crucial for energy metabolism. Prior studies showed that supplemental GAA improved performance in heat-stressed (HS) broilers, but its role in mitigating negative effects in HS pigs remains unexplored. This study evaluated the effect of GAA supplementation on performance and body composition of HS pigs. Eighty gilts (47.6 kg BW; Camborough × AGPIC 337), collectively housed, were assigned to a 2×2 factorial design, with two ambient temperatures (AT; thermoneutral, 22°C; heat stress, 35°C) and two dietary GAA levels (0 and 0.9 g/kg). Diets were formulated according to Rostagno et al. (2017). Over 35 days, pigs had free access to pelleted feed and water. Individual automatic feeders were used to provide experimental diets and to record individual feed intake. Pigs were weighed at days 0 and 35 to calculate average daily gain (ADG) and gain:feed (G:F). Body composition was assessed via dual-energy X-ray absorptiometry at days 0 and 35. Blood samples from 40 pigs (10/treatment) were collected on days 0, 7, and 35 to analyze serum and plasma metabolites. Data were analyzed using GLIMMIX (SAS) including AT, GAA, and their interactions as fixed effects and BW blocks as random effect. Heat stress negatively affected all performance and body composition variables ($P < 0.05$), except G:F ($P = 0.286$). There was no interaction between GAA and the AT ($P > 0.05$). HS pigs supplemented with GAA had higher ADG (0.889 vs. 0.850 kg; $P = 0.022$), and greater body protein deposition than control group (159.8 vs. 152.2 g/day; $P = 0.006$). There was no effect of GAA on ADFI ($P = 0.178$). Blood parameters were similar between treatments except for lactate dehydrogenase, increased by HS but reduced by GAA addition (592.7 vs. 435.0 U/L; $P = 0.049$). These findings suggest GAA supplementation can improve growth performance and protein deposition of HS pigs during the growing phase. **Key Words:** GAA, high ambient temperature, nutritional strategy

**106 Zinc and copper sources as alternatives to pharmacological zinc oxide: impact on growth performance, digestive function and intestinal health.** Hadhemi Baccouri[1], Clara Negrini[2], Alessandra Rigo Monteiro*[3], Luca Lo Verso[1], Marie-Pierre Létourneau-Montminy[1], and Frédéric Guay[1], [1]*Laval University, Quebec, Canada*, [2]*University of Bologna, Bologna, Italy*, [3]*Animine Precision Minerals, Annecy, France*. This project aimed to evaluate the effects of different sources of Zn and Cu on growth performance, microbiota, and intestinal health of weaned piglets. At weaning (d 21), 120 piglets were distributed into 24 pens (5 piglets per pen) and they received one of the 4 treatments: 1) Control: ZnO and $CuSO_4$ (150 mg/kg for both), 2) ZnO+: Pharmacological doses of ZnO and $CuSO_4$ (2,500 and 150 mg/kg), 3) pZn150: Potentiated ZnO (pZn) and monovalent Cu (150 mg/kg for both, Animine, France) and 4) pZn300: pZn (300 mg/kg) and monovalent Cu (150 mg/kg). Experimental diets were provided for the first 14d (phase 1). All piglets received the same standard diets during phases 2 (14–28d) and 3 (28–42d). Piglets were weighed at weaning and on 14, 28, and 42d. Blood samples (d-7 and d-14) and fecal samples (d-14) were collected. The MIXED procedure function was used in fitted linear model and ANOVA. During phase 1, the ZnO+ showed higher average daily gain (ADG) and average daily feed intake (ADFI, $P < 0.01$) than other treatments. For the overall period (days 1 to 42), ADG and ADFI were higher for the ZnO+ and pZn300 compared to the Control ($P < 0.001$). Regarding health markers, D-lactate concentration was lower (P = 0.015) in the pZn300, suggesting improved intestinal permeability. However, analysis of diamine oxidase and citrulline did not show indications of improved intestinal health. Alpha diversity was not affected by diet. In contrast, for beta diversity, the Adonis test showed a significant effect on bacterial structure ($R^2 = 0.21$, P = 0.001), with the ZnO+ displaying a distinct microbiota organization. Supplementation with pZn and monovalent Cu at 300 and 150 mg/kg in the post weaning phase (42d after weaning) resulted in growth performances equivalent to those achieved with ZnO+. However, the benefits cannot be directly related to intestinal markers and microbiota. **Key Words:** nursery piglets, trace minerals

**107 The response of piglets on threonine supplementation to a low threonine diet, in function of dietary protein content.** S. Millet* and S. Goethals, *ILVO (Flanders Research Institute for Agriculture, Fisheries and Food), Merelbeke-Melle, Belgium*. With decreasing dietary crude protein (CP) levels, understanding amino acid (AA) requirements in piglets is crucial. To ensure that the AA of interest is the primary limiting factor in dose-response trials, very low CP levels are often used, which may hinder interpretation if protein or nitrogen limits performance. As a concept, this study tested the effect of supplementing Thr to a Thr-deficient diet (6.7 g/kg SID Thr, 11.5 g/kg SID Lys) at two CP levels (180 g/kg vs. 160 g/kg) in pigs aged 5 to 9 weeks. We hypothesized that a Thr response would depend on the CP level. Pigs were weaned at 4 weeks, and tested between 5 and 9 weeks of age, with 12 pens (6 with barrows, 6 with gilts) of 5 animals per treatment in a RCB design. The 7 treatment groups were: 1) low Thr-high CP; 2) 1+1g Thr; 3) 1+7g urea; 4) low Thr-low CP; 5) 4+1g Thr; 6) 4+7g urea; 7) 4+1g Thr+7g urea. Daily feed intake (g/d) was 567±56, 571±58, 557±52, 555±67, 573±41, 554±63, 554±60; daily gain (g/d) was 417±41, 429±45, 426±45, 388±36, 404±34, 406±44, 415±46; Gain-to-feed (g/kg) was 735±29, 750±14, 766±45, 703±37, 706±33, 734±42, 749±35 for group 1-7. Although the results followed the expected trend, adding Thr did not result in a significant performance improvement, nor was there a significant interaction between CP level and Thr level (P > 0.05). Feed efficiency increased linearly with increased nitrogen content ($P < 0.001$). When analyzing serum urea level as a marker for nitrogen efficiency, the CP:Lys level and Thr in the feed interacted ($P = 0.011$), with $5.5\pm1.7^{ab}$, $4.8\pm3.4^{ab}$, $14.1\pm3.7^{c}$, $3.5\pm1.9^{a}$, $6.6\pm6.3^{ab}$, $13.6\pm5.4^{c}$, $9.6\pm2.6^{bc}$ mg/dl for group 1-7. Adding Thr to a low CP diet seemed to increase serum urea in the low CP diet and decrease in the high CP diet, although no clear distinction was possible with post hoc tests. These results support our hypothesis that nitrogen content may limit performance and that Thr addition only improves nitrogen efficiency when the CP level is high enough. **Key Words:** LYS:CP, amino acid requirement, nitrogen

**108 Increasing exogenous phytase increases phosphorus digestibility but decreases hindgut digestion in phosphorus-deficient diets fed to growing pigs.** J. Y. Sung* and O. Adeola, *Purdue University, West Lafayette, IN, USA.* Eighteen T-cannulated barrows (initial body weight = 52.1 ± 3.0 kg) were assigned to three corn-soybean meal-based diets without inorganic phosphorus supplements, containing 0, 500, or 5,000 FYT/kg of exogenous phytase to investigate the effects of exogenous phytase concentration on hindgut digestion of phosphorus (P), calcium (Ca), and nitrogen. The diets were formulated to contain 1.4 g/kg of standardized total tract digestible P, 3.0 g/kg of Ca, and 162 g/kg of crude protein. Dietary Ca to standardized total tract digestible P was 2.16. Chromic oxide was supplemented at 5 g/kg in each diet as an indigestible marker. The experimental design was a randomized complete block design with body weight as a blocking factor. Daily feed allowance was provided at 3.5% of mean body weight of each block, equally divided into 2 meals, and provided to pigs twice daily at 0800 and 1700 h. Each experimental period consisted of 5-d adaptation, 2-d feces collection, and 2-d ileal digesta collection periods. A polynomial contrast was conducted to determine linear effects of increasing phytase concentration (0, 500, or 5,000 FYT/kg). The apparent ileal digestibility (AID) and apparent total tract digestibility (ATTD) of P linearly increased (24.1 to 67.0%; $P < 0.05$), whereas the hindgut disappearance of P linearly decreased (22.6 to 3.0%; $P < 0.05$) with increasing phytase concentration. As phytase concentration increased, the AID (60.9 to 79.6%) and ATTD of Ca (66.1 to 80.9%) also increased linearly ($P < 0.05$), but there was no significant difference in hindgut disappearance of Ca. The increase in dietary phytase concentration did not affect nitrogen digestion at the ileal, total tract, and hindgut levels. In conclusion, the supplementation of exogenous phytase increased P digestibility but decreased hindgut digestion in P-deficient diets fed to growing pigs. **Key Words:** hindgut, phosphorus, phytase

**109 Effect of dietary benzoic acid on nutrient digestibility and growth performance of weaned pigs fed diets containing pulse grain.** L.F. Wang, E. Beltranena, and R.T. Zijlstra*, *University of Alberta, Edmonton, AB, Canada*. Lentil is a pulse grain that can serve as an alternative feed resource for pigs but may have lower nutrient digestibility than cereal grains. Dietary organic acids such as benzoic acid (BA) may enhance protein and energy digestibility and thereby increase nutrient efficiency and growth performance of weaned pigs. The objective was to test such effects of dietary BA on weaned pigs fed diets with or without lentil. In total, 192 pigs (initial body weight [BW], 8.8 kg) in 12 blocks were fed 4 wheat-canola expeller-soybean meal diets containing either or not lentil in substitution of wheat grain at 20% for phase 1 (day 0–14) and at 30% for phase 2 (day 14–28), either with or without 0.5% BA starting 1 week after weaning. Diets were formulated to provide 10.25 and 10.16 MJ net energy (NE)/kg and 1.32 and 1.22 g standardized ileal digestible lysine/MJ NE for phases 1 and 2, respectively. Data were analyzed as a 2 × 2 factorial arrangement using PROC GLIMMIX of SAS. Dietary lentil decreased ($P < 0.05$) apparent total tract digestibility (ATTD) of dry matter (DM) and gross energy (GE) for phase-1 and phase-2 diets and decreased ($P < 0.05$) ATTD of crude protein (CP) for phase-1 diets. In phase 2, dietary lentil did not consistently decrease diet ATTD of CP. Dietary BA increased ($P < 0.05$) ATTD of DM and GE for phase-1 and phase-2 diets and increased ($P < 0.05$) ATTD of CP for phase-1 diets. In phase 2, dietary BA increased ($P < 0.05$) ATTD of CP for diets without dietary lentil but not for diets with lentil. Feeding lentil decreased ($P < 0.05$) average daily feed intake (ADFI) and average daily gain (ADG) and gain-to-feed (G:F) for diets without BA. Dietary BA increased ($P < 0.05$) ADFI, ADG, and G:F. Feeding lentil decreased ($P < 0.05$) final BW by 0.7 kg and dietary BA increased ($P < 0.05$) final BW by 1.4 kg. In conclusion, 0.5% dietary BA as a functional ingredient increased nutrient efficiency and especially growth performance of weaned pigs fed diets containing pulse grain. **Key Words:** benzoic acid, digestibility, pulse grain

**110 Evaluation of the effect of feeding medium-chain fatty acids on production performance of piglets: a meta-analysis.** A Kihal*, M Puyalto, and JJ Mallo, *Norel Animal Nutrition, Madrid, Spain*. This meta-analysis aimed to evaluate the effect of feeding medium-chain fatty acids (MCFA) on the production performance of piglets. A literature search was

conducted to identify in vivo research studies on MCFA supplementation in piglets. Inclusion criteria were: in vivo, piglets, MCFA, age of animals and doses of each treatment. Eight experiments including 20 treatments and 1,944 animals were selected for this meta-analysis from published papers and congress communications. Data extracted included the number of animals in control and treatment groups and measures of variance of responses (standard error of the mean or standard deviation). The response variables were the production performance of animals: body weight (BW), average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR). A random effect model was used to examine the raw mean difference and standardized mean difference (SMD) between MCFA supplementation and the control group. The analysis was conducted using the metafor package of R. Results of this meta-analysis showed that MCFA considerably improved the production performance of piglets from moderate (< 0.8) to high SMD effect (> 0.8), with high heterogeneity ($50 < I^2 < 60\%$, $P < 0.05$) for FCR and ADFI, to no evidence of heterogeneity ($I^2 = 0.0\%$, $P < 0.05$) for BW and ADFI. Final BW and ADG were increased by 600 g and 24 g/day, or SMD = 0.51 and 0.71 ($P < 0.05$), respectively. The FCR was ameliorated for treatment groups compared to the control group by 0.08 units or with an SMD = 1.23 ($P < 0.05$). Meanwhile, the ADFI of pigs was not affected by the treatment in comparison to the control group ($P > 0.05$). In conclusion, MCFA are bioactive lipids recommended to be an efficient strategy to enhance the intestinal health of piglets and their production performance. **Key Words:** medium-chain fatty acids, piglet, intestinal health

**111 High dietary lysine to enhance growth following a dietary protein restriction after weaning.** M. Girard[2], G. Bee[2], P. Silacci[2], R. Ruggeri[2], C. Larsen[1], and J.G. Madsen*[1], [1]*Department of Veterinary and Animal Sciences, University of Copenhagen, 1870 Frederiksberg, Denmark*, [2]*Research Group Swine Nutrition and Pork Quality, Agroscope, 1725 Posieux, Switzerland*. Compensatory growth might counter impaired growth resulting from pigs being fed a low-protein (LP) diet during early weaning. It was hypothesized that high dietary lysine (Lys) in a 4-week (W) recovery period (W5-8) after a 4-W protein restriction period (W1-4) could enhance compensatory growth. 60 piglets from 20 litters of 1 farrowing series were categorized being average growers (200-220 g/day) in the pre-weaning period. On day of weaning pigs were pre-allocated to 1 of 4 dietary treatments (DT): control (Ctrl, 154.8 g dCP, 10.17 g SID Lys per kg), standard (ST, 154.8 g dCP, 10.17 g SID Lys per kg), ST+10% Lys (Lys10, 154.2 g dCP, 11.19 g SID Lys per kg) and ST+20% Lys (Lys20, 155.7 g dCP, 12.21 g SID Lys per kg). From weaning to W4 after weaning (housed 6 pigs/pen) ST, Lys10 and Lys20 pigs were fed the same LP (122.9 g dCP/kg, 10.17 SID g Lys/kg) starter diet, whereas Ctrl pigs were fed a diet formulated according to recommendations. From W5 to W8 after weaning (housed 4 pigs/pen) ST pigs were fed a diet with standard protein level, and Lys10 and Lys20, respectively, fed a diet with ST protein level plus 10% and 20% Lys including remaining EAA. Data were analyzed using ANOVA function in R, with DT and W and their interaction as fixed effects. No difference ($P > 0.05$) in BW and ADG between pigs fed Ctrl and LP diet in W1-4, but LP pigs displayed greater ($P < 0.05$) total FI per pig (11.4 vs. 10.8 kg) and greater ($P < 0.05$) ADFI per pig (0.42 vs. 0.40 kg/d) in the same period. After the restriction period Ctrl and Lys20 pigs did not differ ($P > 0.05$) in ADG in W7 (1.11 vs. 0.96 kg/d) and W8 (1.01 vs. 0.95 kg/d) nor FI in W5-8, but ST displayed lower ($P < 0.05$) ADG than Ctrl (0.91 vs. 1.11 kg/d) in W7 but not W8 ($P > 0.05$, 0.85 vs. 1.01 kg/d) and lower ($P < 0.05$) ADFI (1.3 vs. 1.5 kg/d) in W5-8. In conclusion, pigs fed LP diet compensated by increasing FI in the restricted period, while Lys20% compared with Ctrl pigs maintained ADG throughout the recovery period. **Key Words:** Compensatory growth, lysine, body composition.

## Theme II: Advances in understanding of nutrient digestion and absorption

**112 Modulators of passage and nutrient absorption kinetics in the digestive tract of pigs.** S de Vries* and WJJ Gerrits, *Animal Nutrition Group, Wageningen University & Research, Wageningen , the Netherlands*. The shift in available resources for pig feeds challenges traditional feed evaluation approaches. Static models, with fixed input variables per ingredient and assuming additivity, no longer suffice to account for the dynamic nature of future raw material supplies. To address continuous variation in ingredient quality, insights into the relation between nutrient supply and metabolic fate in the animal become increasingly important. Dynamic feed evaluation models and precision feeding techniques aimed at accurately matching supply with requirements for groups of animals considering factors like age, health status, genetic potential, and production system, can facilitate optimal use of resources, animal performance, and health. A key element of such models is the amount of nutrients available to the animal throughout the day, which requires understanding of the kinetics of nutrient degradation and absorption in the gastrointestinal tract. Besides intrinsic ingredient properties and animal factors, such as feed intake patterns, physicochemical properties of the diet are important modulators. Results of recent studies on nutritional regulation of digestion and absorption kinetics will be presented and discussed. Particular focus is placed on how dietary fibres regulate digesta passage and nutrient solubilization kinetics, depending on fibre type and diet properties such as particle size and gelling behaviour, and their measurement *in vitro* and *in vivo*. Beyond their impact on large intestine passage and fermentation kinetics, fibres are key regulators of gastric emptying, thereby modulating the timing of nutrient delivery into the small intestine, while also affecting intragastric mixing and acidification kinetics. These insights enhance understanding of the digestion process and thereby aid in optimizing feeding strategies with diets containing increasing quantities of fibre-rich byproducts. **Key Words:** fibres, digestion, fermentation

**113 *In vitro* fermentation potential of undigested dietary protein in growing pigs.** H. Zhang*[1,2], J. Cone[1], A.K. Kies[3], W.H. Hendriks[1], and N. van der Wielen[1,4], [1]*Animal Nutrition Group, Department of Animal Sciences, Wageningen University & Research, Wageningen, The Netherlands*, [2]*State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China*, [3]*ArieKiesAdvies, Druten, The Netherlands*, [4]*Division of Human Nutrition and Health, Department of Agrotechnology and Food Sciences, Wageningen University & Research, Wageningen, The Netherlands*. Undigested dietary proteins entering the hindgut play a pivotal role in microbial fermentation processes, influencing gut health and nutrient utilization in pigs. This study assessed the *in vitro* fermentation potential of ileal digesta from pigs fed diets containing one of seven protein sources as the sole dietary protein: maize germ meal (MGM), cottonseed meal (CSM), rapeseed cake (RSC), rapeseed meal (RSM), peanut meal (PM), soybean meal (SBM), and sunflower meal (SFM). Digesta samples (10 mg nitrogen each) were incubated with faecal inoculum derived from 20 pigs fed commercial diets under standardized anaerobic conditions. Whey protein isolate (WPI), included without prior digestion, served as a control. Protein utilization was assessed via gas production (GP) over 48 hours, analysed through a biphasic model to delineate substrate fermentation and microbial turnover dynamics. Statistical analysis was conducted in SAS, with Dunnett's test used to compare ileal digesta against WPI and Tukey's test to evaluate differences among protein sources. Higher maximum GP rates ($R_{max}$) were found in ileal digesta samples compared to WPI ($P < 0.05$). Among protein sources, $R_{max}$ ranged from 16.8 ± 0.6 for MGM to 27.9 ± 0.8 mL/h for SFM, while cumulative GP and turnover slopes further highlighted distinct fermentation characteristics. Solubility and molecular mass analyses revealed variations in the proportions of insoluble nitrogenous molecules and the molecular mass distribution of soluble molecules, highlighting differences in fermentability among protein sources. The calculated standardized hindgut fermentation potential ranked as follows: SBM<PM<CSM<RSC<SFM<RSM<MGM (0.46, 0.54, 0.61, 0.69, 0.82, 0.83 and 1.18 L/g ingested crude protein, respectively). This study provides a novel perspective on evaluating the potential accessibility of protein sources to be utilized

by hindgut microbiota, offering valuable insights into optimizing feed formulations to enhance gut health in pigs. **Key Words:** *In vitro* fermentation, Protein, Ileal digesta

**114 Evaluation of soybean-derived trypsin inhibitor proteins on gastric emptying, luminal pH, and endogenous enzyme activity in late-stage nursery pigs.** MJ Nisley*[1], ER Burrough[1], HB Krishnan[2], and NK Gabler[1], [1]*Iowa State University, Ames, IA, USA*, [2]*University of Missouri, Columbia, MO, USA*. Trypsin inhibitor proteins, intrinsic to soybeans, are known to attenuate pig growth and protein digestion efficiency while upregulating endogenous protease secretion. The objective of this research was to evaluate how soybean-derived dietary active trypsin inhibitor units (TIU) would impact gastric emptying, luminal pH, and endogenous enzyme activity in late-stage nursery pigs. Sixteen pigs (5.6 ± 0.90 kg BW) were allotted across two diet treatments consisting of 1) Low TIU (CON, 0.87 TIU $mg^{-1}$) or 2) high TIU (HIGH, 4.29 TIU $mg^{-1}$). On d 35, pigs were fasted overnight, given an *ad libitum* meal, and refusals were recorded after 30 min. Pigs were sacrificed 240 min postprandial to evaluate stomach meal retention and gastric emptying rates. Stomach, ileal, cecal, and colonic contents were collected and analyzed for pH, and ileal content trypsin activity was measured. Data were analyzed using a t-test with the pig as the experimental unit. Compared to the CON pigs, HIGH pigs observed reduced final BW (21.6 vs 18.0 kg, $P = 0.007$), weight gain (0.46 vs 0.36 kg $d^{-1}$, $P = 0.003$), and feed intake (0.70 vs 0.57 kg $d^{-1}$, $P = 0.002$). Following fasting, gastric emptying rate (dry matter, DM) was slower in CON pigs (2.11 g $h^{-1}$ kg $BW^{-1}$) compared to HIGH pigs (2.74 g $h^{-1}$ kg $BW^{-1}$; $P = 0.028$). However, meal feed intake (DM) was similar in CON pigs (12.2 g kg $BW^{-1}$) and HIGH pigs (13.5 g kg $BW^{-1}$; $P = 0.263$). Stomach pH was higher in CON pigs compared to HIGH pigs (2.87 vs. 2.16, respectively; $P = 0.033$). No significant differences were observed in ileal and cecal pH between treatments, but colonic pH was higher in CON pigs compared to HIGH pigs (6.25 vs. 5.86, respectively; $P = 0.040$). Compared to CON pigs, ileal endogenous trypsin activity was potentiated in HIGH pigs (66 vs. 556 mU $mL^{-1}$, respectively; $P = 0.006$). In conclusion, high TIU diets attenuate pig growth and digestion efficiency while modulating gastric emptying, altering intestinal pH, and upregulating endogenous trypsin secretion. **Key Words:** Trypsin gastric digestion

**115 Basal ileal endogenous crude protein and amino acid losses in swine is influenced by age.** JAL Barbosa*[1], H Moreira Junior[1], JL Brito[1], CEM Bertanha[1], SSS Sousa[1], A Gorrosterrazú[1], MLP Tsé[2], ABS Oliveira[3], F Dilelis[1], and US Ruiz[1], [1]*University of São Paulo (USP), Luiz de Queiroz College of Agriculture, Department of Animal Science, Piracicaba, São Paulo, Brazil*, [2]*São Paulo State University (UNESP), School of Veterinary Medicine and Animal Science, Department of Animal Production, Botucatu, São Paulo, Brazil*, [3]*Ingredion, Mogi Guacu, São Paulo, Brazil*. The determination of standardized ileal digestibility of crude protein (CP) and amino acids (AA) of feed ingredients in pigs involves the quantification of basal endogenous losses (BEL) of CP and AA in ileal digesta, derived from digestive secretions, such as enzymes and mucoproteins, and desquamated cells from the intestinal epithelium. The age or category of the pig may influence BEL in pigs. Therefore, the objective of this study was to evaluate the BEL of CP and AA in female pigs at four different categories: 28 days (7.80 ± 0.55 kg body weight - BW), 78 days (33.99 ± 3.02 kg BW), 125 days (79.82 ± 4.05 kg BW), and 243 days (169.03 ± 6.50 kg BW). Twenty-four female pigs, 6 animals per category, were fed a nitrogen-free (NF) diet containing starch, sugar, cellulose, oil, minerals, vitamins, and 0.5% titanium dioxide, used as an indigestible marker. The animals were fed at 2.8 × DE for maintenance for 14 days, and on the 15th day, the pigs were euthanized for ileal digesta collection and determination of CP and AA contents, which were expressed as g/kg of dry matter (DM) feed intake. A randomized block design was adopted, with 4 treatments (animal ages), 6 repetitions per treatment, and 1 animal per experimental unit. The data were subjected to analysis of variance using the SAS MIXED procedure and regression analysis. Linear increases ($P < 0.05$) in the BEL of Lys (0.37 to 0.68 g/kg DM) and His (0.10 to 0.40 g/kg DM) were observed, and trends for linear increases of Ala ($P = 0.094$) and Ser ($P = 0.076$) were verified. The BEL of CP, Arg, Leu, Gly and Pro had a

quadratic effect ($P < 0.05$), with the highest values observed in the pigs at 125 days of age (CP = 30.85 g/kg DM; Arg = 0.93 g/kg DM; Leu = 0.88 g/kg DM; Gly = 4.17 g/kg DM; Pro = 5.43 g/kg DM). The total AA BEL also presented a quadratic effect, but the highest values were observed in the pigs at 78 days of age (Total AA = 23.48 g/kg DM). In conclusion, the age of pigs is an important factor on the BEL of CP and AA. **Key Words:** Basal Endogenous Losses, Ileal Digestibility, Age of Pigs

**116 Feasibility of using an x-ray fluorescence device for digestibility studies in pigs.** Y.J.Y. Manaig*[1], E. Gourlez[2], M. Taris[1], A.R. Monteiro[1], and F. De Quelen[2], [1]*Animine, Annecy, France*, [2]*INRAE, Institut Agro Rennes-Angers, PEGASE, Saint Gilles, France*. AniGun®, a portable X-ray Fluorescence (XRF) device (Animine, France), has been calibrated for rapid, cost-effective mineral analysis of forages for precision ruminant nutrition. This study evaluated the feasibility of using portable XRF for calibrating and analyzing the mineral composition of swine feces. It offers a potentially cheaper and simpler alternative to the widely accepted inductively coupled plasma-optical emission spectroscopy (ICP-OES) method for measuring trace mineral concentration in digestibility studies. Fecal samples from a previous trial, where 70 pigs were fed varying levels of Cu and Zn, were lyophilized and analyzed for minerals (Cu, Zn, Mn, Ca, K, S, P) using ICP-OES. Thirteen fecal samples representing the highest, lowest, and median values for each mineral were selected to build a multi-elemental calibration, covering ranges: 30-210 mg/kg for Cu; 144-849 mg/kg for Zn; 305-523 mg/kg for Mn; 9.2-16.9 g/kg for Ca; 3.1-7.8 g/kg for K; 1.1-1.9 g/kg for S; and 4.6-8.8 g/kg for P. Using these data, linearity, limits of detection, and quantification were established for each element. To validate the calibration, accuracy and precision of the XRF measurements were evaluated using random samples from the same pool. Agreement between portable XRF and ICP-OES was assessed through mean percentage bias and recovery, with results indicating mean percentage recoveries of 97.4% for Cu, 93.9% for Zn, 96.5% for Mn, 99.2% for Ca, 95.8% for K, 94% for S, and 95.6% for P, with a mean bias of ~4%. In summary, these findings demonstrate the potential of this portable XRF as a valuable tool for advancing the understanding of trace mineral digestion and excretion in swine. With the device's efficiency, it offers an effective alternative in analyzing mineral concentration for digestibility and performance trials. Future research should explore expanded calibrations and applications to tissues, feed, and feed ingredients, contributing to digestibility studies. **Key Words:** digestibility, feces, XRF

**117 Fiber Fermentation Kinetics of Wheat and Maize in Weaned Piglets.** I. Kaikat*[1], L. Blavi[2], M. A. Ton Nu[2], S. Tibble[2], A. Koppenol[2], G. González-Ortiz[3], and J. F. Pérez[1], [1]*Animal Nutrition and Welfare Service (SNiBA), Department of Animal and Food Science, Universitat Autònoma de Barcelona (UAB), 08193 Bellaterra, Spain*, [2]*AB Neo, PL Fraga, C/ Comunidad de Murcia, parc. LIE-1-03, 22520 Fraga (Huesca), Spain*, [3]*AB Vista, Marlborough SN8 4AN, United Kingdom*. Understanding the site and extent of fiber fermentation in the digestive tract can influence cereal selection in feed formulation to improve gut health in weaned piglets. Twenty-four weaned piglets (21d, 5.6 ± 0.84 kg) were used to compare fiber fermentation kinetics of wheat and maize. Piglets were divided into 8 pens (4 pens/diet, 3 pigs/pen) and fed an adaptation diet in week 1 and the 2 experimental diets with 91.6% wheat or 90.6% maize in week 2. Ytterbium oxide (50mg/kg) was used as an inert marker. On d15, ileal and proximal colon digesta, and fecal samples were collected for the analyses of starch, non-starch polysaccharides (NSP) and volatile fatty acids (VFA). Total NSP (tNSP) in cereal was classified based on its fermentation site as fast (fNSP – proximal colon), slow (sNSP – lower colon) and resistant NSP (rNSP – feces). Data were analyzed with cereal type as fixed effect and pen as random effect. Higher tNPS was observed in wheat compared to maize (76.70 vs 59.06 g/kg) due to higher soluble NSP (19.74 vs. 5.13 g/kg). Total tract tNSP fermentation was 39% in wheat and 35% in maize. Wheat and maize had 34 and 23% fNSP, 5 and 11% sNSP, 61 and 65% rNSP of tNSP, respectively. Within fNSP, the soluble fraction was higher in wheat than maize (12.2 vs. 1.6 g/kg DM, $P = 0.003$). Wheat had a higher starch content (606 g/kg) compared to maize (557 g/kg) and total tract digestibility of starch was significantly higher in wheat than in

maize (99.7% vs. 98%; $P$ = 0.013). No differences were observed on the fermentation metabolites production in feces, but in the proximal colon, wheat-fed piglets tended to show higher VFA:BCFA (branched-chain fatty acids) ratio than maize-fed piglets (153.9 vs. 67.3; $P$ = 0.059). Wheat results in higher carbohydrate fermentation in both the upper and lower gut compared to maize, promoting fiber utilization and gut microbial activity. These findings highlight the potential of wheat-based diets to improve gut function in weaned piglets. **Key Words:** Non-starch polysaccharides, Fermentation, Weaned piglets

## Theme III: Functional Ingredients and Utilization of Feed Resources for Improved Digestive Function and Nutrient Efficiency

**118 Approaches for reducing nitrogenous/phosphorus waste excretion in the pig - challenges and opportunities.** Léa Cappelaere[1,2], Florence Garcia-Launay[3], Patrick Schlegel[2], and Marie-Pierre Létourneau Montminy*[1], [1]*Laval University, Quebec, Quebec, Canada*, [2]*Agroscope, Posieux, Switzerland*, [3]*INRAE UMR PEGASE, Saint-Gilles, Britanny, France*. The intensification of agricultural systems has increased yields and efficiencies, reducing environmental impacts and production costs. However, nutrient cycles have become dissociated in many agricultural systems, including swine production systems. Losses of nitrogen (N) and phosphorus (P) from manure lead to severe eutrophication and acidification problems in many regions of the world, as well as greenhouse gases emissions; but they are also essential nutrients for crop production. In a dissociated nutrient cycle, the use of mineral phosphate and urea as fertilizers becomes essential. For animal, P is essential for growth performance and bone mineralization. Additionally, since it is a limited and non-renewable resource produce by few countries, phosphate, dictates our capacity to produce food. Therefore, an efficient use of plant P by pigs and a consequent use of excreted P as fertilizer for crop production is necessary. N is also essential for animal and plant production. N is not a finite resource, but production of mineral N fertilizers requires a high energy input and protein-rich feedstuffs can have a high negative environmental impact depending on production context which requires to reduce N excesses. These environmental challenges are even-more relevant for competitiveness given the higher prices of raw materials and N and P inputs for feedstuffs in recent years. In consequence, there is an urgent need to optimize utilisation of resources such as P and N. To achieve this, an accurate and robust feeding system for P (including calcium (Ca) given their many interactions) and N (including amino acids (AA)) is needed. This involves setting up a system that 1) accurately and robustly determines the P, Ca and AA content of feed ingredients, 2) determines precisely the P, Ca and AA requirements of animals, 3) provides these nutrients to animals as accurately as possible and 4) integrates innovative feeding strategies to optimize their utilization by animals. **Key Words:** Phosphorus, nitrogen, environment

**119 Improving starch digestion in barley, wheat and maize by xylanase/glucanase, phytase, protease and their combination in an *in vitro* digestion model.** X. Liu*[1], B.M. Flanagan[1], E. Roura[1,2], and M.J. Gidley[1], [1]*Centre for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Brisbane, Queensland, Australia*, [2]*Centre for Animal Science, Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Brisbane, Queensland, Australia*. Grains are the primary source of energy in pig diets. This study investigated the effects of commercial exogenous enzyme feed additives on starch digestion in hammer-milled barley, wheat, and maize grains, using an *in vitro* digestion model. A 5 × 3 factorial design was used to test five enzyme treatments (control, phytase, xylanase/glucanase, protease, and Combo - a combination of all enzymes) across the three grains. The digestion model was adapted from the standard Infogest 2.0 method, which simulates oral (2 min, pH 7), gastric (2 hr, pH 3), and small intestinal (8 hr, pH 7) digestion. Digesta samples were collected at 0, 20, 40, 60, 120, 180, 240, 360 and 480 mins during small intestinal digestion. Starch digestion was quantified by two parameters: (1) starch digestibility and (2) digestion rate coefficient, which represent how much and how fast starch was digested respectively. The effect of enzyme treatments on starch digestion for each grain was analysed using one-way ANOVA followed by Tukey's post-hoc test. Our results indicate that the enzyme treatments improved starch digestion parameters of the three grains to different extents. In barley, xylanase/glucanase and Combo significantly improved both starch digestibility and digestion rate coefficient ($P < 0.05$) while phytase significantly improved digestion rate coefficient and tended to increase starch digestibility ($p = 0.051$). In wheat, all the enzymes tested significantly improved starch digestibility ($P < 0.05$) and xylanase/glucanase also significantly improved digestion rate coefficient ($P < 0.05$). In contrast, for maize, only phytase and Combo significantly improved both starch digestibility and

digestion rate coefficient ($P < 0.05$). Mechanistic origins for these differential effects of enzymes on grains will be discussed. In conclusion, our results suggest that the selection of enzyme feed additives should be tailored to specific feed ingredients for optimal digestion performance. **Key Words:** xylanase/glucanase, phytase, protease

**120** ***In vitro*** **evaluation of chicory-induced modulation of intestinal health in weaning piglets: Approach combining in vitro digestion, dialysis, and fermentation with a triple cell culture model.** T.S. Kulkarni*[1,2], P. Siegien[2], L. Comer[3], A. Richel[2], B. Cudennec[1], C. Dugardin[1], S. Theysgeur[1], A. Lucau[4], N. Everaert[3], M. Schroyen[2], and R. Ravallec[1], [1]*UMR-T 1158, BioEcoAgro, University of Lille, Lille, FRANCE*, [2]*Precision Livestock and Nutrition Laboratory, TERRA Teaching and Research Centre, Gembloux Agro-Bio Tech, University of Liège, Gembloux, BELGIUM*, [3]*Nutrition and Animal Microbiota EcoSystems lab, Division of A2H, Department of Biosystems, KU Leuven, Leuven, BELGIUM*, [4]*Joint Laboratory CHIC41H University of Lille-Florimond-Desprez, Lille, FRANCE*. Weaning can disrupt the porcine gastrointestinal (GI) tract, leading to chronic inflammation. While *Cichorium intybus*-extracted inulin improves gut health, its extraction is costly & certain bio actives are removed. Therefore, this study aimed to compare the effects of crude chicory (Chi) vs inulin (Inu) on gut health using an *in vitro* digestion-dialysis-fermentation combined with a triple cell culture (Caco2/HT29-MTX/U937). Human cell lines were used to create a more comprehensive model due to similarities between human & piglet GI physiology. The experiment had 5 groups equated (eq.) for 'inulin content' at respective concentrations: Inu, Chi, Feed (F), F+2% Inu (F2I) & F+2% Chi (F2C). After digestion & 24h dialysis, samples were fermented using a weaned piglet faeces inoculum. Gas volume was measured for 48h, & fermentation broth (FB) was collected at 12h. Differentiated Caco2/HT29 in the apical part were pre-treated with LPS (-ctrl) or dexamethasone (+ctrl) 6hr before and during incubation of 12h FB for 24h. Cytokine levels were analyzed in the basolateral media with differentiated U937 (macrophages), gene expression was measured in the apical cells, & permeability by Lucifer Yellow. Inu produced more gas & short chain fatty acids than Chi but F2C had higher levels of butyrate than F2I ($P < 0.0001$). Chi & F2C showed a higher α-diversity (Shannon & Simpson) than Inu eq. & F2I ($P < 0.05$). Compared to Inu, Chi increased the abundance of beneficial microbiota such as *Lactobacillus*, *Bifidobacterium*, *Butyricicoccus* & *Ruminococcus* ($P < 0.0001$). After 24h FB treatment & LPS stimulation, Inu & Chi downregulated pro-inflammatory cytokines IL-1β, IL8, TNF-α & genes MAPK14, MyD88, AKT1 ($P < 0.05$). Interestingly, F2C showed a more significant effect than F2I ($P < 0.05$). Chi had a 3-fold increase in MUC2, while Inu eq. upregulated JAMB expression ($P < 0.05$). F2I had a higher permeability than F2C ($P < 0.05$). In conclusion, cost-effective Chi showed greater benefits in enhancing gut health. **Key Words:** Chicory, in vitro, gut

**121** **Safe level of soy antinutritional factors in diets of weaned piglets.** M. A. Ton Nu*[1,2], L. Blavi Josa[2], L. Sobrevia[2], S. Laird[2], S. Tibble[2], and A. Koppenol[2], [1]*AB Neo a/s, Videbaek, Midtjylland, Denmark*, [2]*AB Neo, Fraga, Huesca, Spain*. The study aimed to evaluate soy antinutritional factors (ANF) effect on performance and gut health of weaned piglets. Weaned piglets (960 in total, 6.93 ± 0.23 kg, n=12) were placed into 4 groups with increasing dietary soy protein (SP) at 0 (Control - 14% fishmeal), 6.5, 13 and 19.5% corresponding to 0, 0.33, 0.65 and 0.98% oligosaccharides (OS) and 0, 0.08, 0.17, 0.25 mg/g trypsin inhibitor activity (TIA), respectively. Pigs were fed 4 diets for 4 weeks and feces were collected from 9 pens/group at D28. Body weight (BW) and feed intake (FI) were recorded at D 14 and 28 while fecal score, survivability, morbidity, and medication were recorded daily. Data were analyzed with fixed effect of treatment and sex and their interaction with BW0 as covariate. No treatment effect was observed on average daily gain (ADG) at D1–14 and BW at D14 ($P > 0.05$). However, 13%-SP had higher FI than control (257 vs, 213 g/d, $P = 0.02$). Control had lower FCR than 13%- and 19.5%-SP (0.91 vs. 1.12, $P < 0.05$). At D28, 6.5% SP had higher BW by 700g than 19.5% SP ($P = 0.046$). On D14–28, no difference in growth between treatments vs. control ($P > 0.05$) except 6.5%-SP had better FCR than control (1.13 vs. 1.25, $P = 0.03$). On D1–28, piglets fed 6.5% SP had better ADG (344 vs. 316 g/d) and better FCR (1.04 vs. 1.15) than 19.5% SP ($P < 0.05$). No

difference was observed between control vs. treatment on growth, fecal score, survivability, morbidity, medication, volatile fatty acids (VFA), short chain fatty acids, branch chain fatty acids (BCFA), VFA:BCFA, *Lactobaccillus* and *E. Coli* in feces ($P > 0.05$). All treatments with SP had higher fecal *Bifidobacterium* than control ($P = 0.001$) which indicated OS prebiotics effect. No linear correlation was observed between increasing dietary SP or ANF and performance or gut health. In conclusion, dietary ANF of up to 0.98% OS and 0.25 mg/g TIA did not negatively influence performance and gut health but promoted beneficial gut microbiota in piglets. **Key Words:** soy oligosaccharides, trypsin inhibitor, gut health

**122 The ratio of cystine to protein as a potential indicator of digestible amino acid concentration in heat-damaged animal byproducts for growing pigs.** J. Y. Sung*[1], M. K. Wiltafsky-Martin[2], and O. Adeola[1], [1]*Purdue University, West Lafayette, IN, USA*, [2]*Evonik Operations GmbH, Hanau, Germany*. Heat damage during the rendering process can potentially reduce both amino acid digestibility and concentration in animal byproducts including poultry meal (PM) and meat and bone meal (MBM). For this reason, different digestible amino acid values should be applied to each PM or MBM in diet formulation based on the severity of heat damage. The hypothesis of the current study was that the ratio of cystine to crude protein (CP) could be a potential indicator of heat damage. Therefore, the objective of the current study was to establish equations for predicting standardized ileal digestible (SID) amino acids in heat-damaged PM and MBM based on the ratio of cystine to CP. Data were collected from two previous studies that reported a linear or quadratic decrease in the standardized ileal digestibility of amino acids in PM and MBM autoclaved at 134°C for 0 to 180 min. Cystine was the most heat-sensitive component in PM (7.1 to 3.5 g/kg) and MBM (4.4 to 1.8 g/kg) after 180-min autoclaving. In contrast, lysine concentration remained relatively constant in PM (42.3 to 38.0 g/kg) and MBM (24.4 to 23.1 g/kg) after 180-min autoclaving. Equations for predicting SID amino acids in PM and MBM were established using the REG procedure of SAS. Equations for estimating SID lysine (g/kg, as-is basis) in heat-damaged PM and MBM meal based on the ratio of cystine to CP (g/kg) are as follows: SID lysine in PM for pigs (31 to 14 g/kg, as-is basis) = –6.92 + 3.65 × cystine to CP ratio, $R^2 = 0.96$; SID lysine in MBM for pigs (19 to 11 g/kg, as-is basis) = –7.85 + 7.09 × cystine to CP ratio – 0.45 × (cystine to CP ratio)$^2$, $R^2 = 0.93$. The ratio of cystine to CP was also highly associated with SID amino acids in PM ($R^2 = 0.95$ to 0.97) and MBM ($R^2 = 0.89$ to 0.93). In conclusion, cystine was the most heat-sensitive component in heat-damaged animal byproducts and the ratio of cystine to CP could be used to estimate digestible amino acid concentration in heat-damaged PM and MBM for growing pigs. **Key Words:** Animal byproduct, cystine to protein ratio, digestible amino acid

**123 Effect of bakery products and legume seeds in the diet on nutrient digestibility of growing-finishing pigs.** M. van Helvoort*[1] and P. Bikker[2], [1]*De Heus Animal Nutrition, Ede, The Netherlands*, [2]*Wageningen University & Research, Wageningen Livestock Research, Wageningen, The Netherlands*. Sustainability of pig production can be enhanced by its integration in a circular food system. Circularity can be improved by replacing dietary cereal grains and soybean meal (SBM) with former food and legume seeds (LS). This study was conducted to evaluate the linear and quadratic effects of replacing SBM, palm kernel expeller (PKE) and cereal grains with bakery products (BP) and LS on nutrient digestibility in growing-finishing pigs. A control diet high in cereal grains, with SBM and PKE was replaced with a diet high in BP with approximately 50% bread meal and a diet high in LS with 22.5% peas and 25% faba beans, both in 3 equidistant steps. A total of 736 pigs (29-124 kg) were randomly assigned to 16 replicate pens. Faecal grab samples were collected from 4 pigs per pen for 3 consecutive days during week 6. Apparent total tract digestibility (ATTD) was determined using acid insoluble ash as marker. Regression analysis was used to quantify the impact of dietary BP or LS content on nutrient digestibility. Limited effects were found on growth rate and feed efficiency. ATTD of dry matter (DM), organic matter (OM), crude protein (CP), crude fat (CF) and non-starch polysaccharides (NSP) of the control diet was 81.4%, 83.9%, 70.9%, 73.4% and 55.2%. Results indicated significant linear, but no quadratic effects of BP and LS content on nutrient digestibility. Complete replacement by

BP decreased ATTD of DM, OM and NSP by 2.8%, 2.5% and 1.5%, respectively ($P \leq 0.05$), while increasing ATTD of CF by 4.9% ($P \leq 0.05$). Complete replacement by LS reduced ATTD of DM, OM and NSP by 5.3%, 6.4% and 10.1%, respectively ($P \leq 0.05$). Neither BP nor LS replacement significantly affected ATTD of CP ($P \geq 0.10$). Comparison of ATTD with Dutch CVB feed table values showed good predictions for the control and BP diets, whereas the LS diet had slightly lower ATTD than CVB values. In conclusion, replacing cereal grains, SBM and PKE with BP and LS linearly reduced ATTD of DM, OM and NSP, but not of CP. **Key Words:** circularity, former food, legume seeds

**124 A new sustainable grain protein concentrate can replace soy protein concentrate or hydrolyzed wheat gluten in piglet diets.** L.C.M. van Enckevort*, P.T. van 't Veld, and I.M. van As, *Denkavit Netherlands B.V., Voorthuizen, Netherlands.* To study if a newly developed Grain Protein Concentrate (GPC, 78% cp) derived from grain side-streams can replace soy protein concentrate (SPC) or hydrolyzed wheat gluten (HWG) in piglet diets, three trials were conducted. In the first trial (T1) a total of 414 piglets (23 replicates per treatment, 6 piglets per pen) were fed from 0-15 days post weaning a control diet with 5% SPC (A), or with 50% (B) or 100% replacement (C) of SPC by GPC. In the 2nd trial (T2) with 20, 20 and 18 replications (litters) respectively, the effect of providing a creep feed from day 5 to 22 of age with 5% SPC (A), or with 50 % (B) or 100% replacement (C) of SPC by GPC was measured. In the third trial (T3) with 300 piglets (25 replicates per treatment, 6 piglets/pen), the performance was measured of piglets fed a weaner diet with 2.5% GPC (A) or 2.4% HWG (B).
Data was statistically analyzed with ANCOVA model using weaning weight as co-variable in trial 1 and 3 and birth weight in trial 2. In T1 from 0-15 days post weaning, weight gain (2.7 vs. 3.4 vs. 3.4 kg/piglet; $P < 0.001$), feed intake (3.7 vs. 4.1 vs. 4.0 kg/piglet; $P = 0.03$) and feed efficiency (0.74 vs 0.84 vs 0.84: $P < 0.001$) was higher in group B and C than group A. In T2 the creep feed intake of suckling piglets from day 5 to 22 (104 vs 122 vs 128 g/piglet) in group B and C tended to be higher ($P < 0.10$) than in group A. Mortality (1.1 vs 0.5 vs 0.4 piglets per litter) in group B and C was significantly ($P < 0.05$) lower than in group A. In T3, the weight gain, feed intake and feed efficiency in group A and B were not significantly different. The calculated Carbon Food Print, using PEFCR-method and GFLI-database, of the diet with 5% GPC as replacement of SPC in T1 and T2 was reduced with 22%. A newly developed Grain Protein Concentrate can replace soy protein concentrate or hydrolyzed wheat gluten in piglet diets in a much more sustainable way, without compromising performance. **Key Words:** sustainable, protein concentrate, piglet

**125 *Probiotic Bacillus subtilis* C-3102 improved sow performance and reduced scouring on its progeny.** JB Lacuesta*[1], E Angeles[1], JM Raquipo[1], KJ Gayosa[1], and R Masilungan[2], [1]*Philchema, Inc, Quezon City, Philippines*, [2]*Phillippines College of Swine Practitioners, Quezon City, Philippines*. The probiotic, *Bacillus subtilis* C-3102 in swine has the potential on improving gut health, immunity and performance that enhances nutrient digestion, and scouring and antibiotic usage reduction. The study was done to determine the effects of probiotic, *Bacillus subtilis* C-3102 on sow reproductive parameters and scouring on its progeny. A total of 20 crossbred female sows (average parity: 5±2.10) were randomly assigned to two dietary treatments following a randomized complete block design, with parity as the blocking factor. Treatments were, Treatment 1 (T1) – lactating diet without probiotic (n = 10) and Treatment 2 – T1 added with probiotic, *Bacillus subtilis* C-3102 at 1.0 kg/ton (n = 10). Lactating diet has 2,565 kcals/kg net energy and 1.25% standardized ileal digestible lysine. Feeding was at day 84 of gestation to weaning. Piglets produced per treatment were assigned to the same experimental group. Treatments were, Treatment 1 - piglets fed diets with 0.5 kg/ton Colistin (n = 109) and Treatment 2 – full replacement of Colistin with probiotic, *Bacillus subtilis* C-3102 at 0.5 kg/ton (n=107). Creep feed contained 2,554 kcal/kg net energy and 1.30% standardized ileal digestible lysine. Feeding was from birth to post-weaning. Birth weight were gathered at farrowing, and scouring at pre-weaning and post-weaning. Data gathered were analyzed using Student T-test of Microsoft Excel. The α-level used to determine the significance and tendencies between means were 0.05 and 0.10, respectively. Probiotic, *Bacillus*

*subtilis* C-3102 supplementation increased birth weight ($P$ = 0.005) and decreased scouring during the pre-weaning phase ($P$ = 0.038) and tended to decrease during the post-weaning phase ($P$ = 0.072). No treatment differences ($P$ > 0.05) were found on the other parameters measured. The results suggest that the effects of supplementing probiotic, *Bacillus subtilis* C-3102 in sows improving birth weight and offers an alternative to Colistin in addressing scouring in piglets. **Key Words:** : *Bacillus subtilis* C-3102, reproductive, scouring

**126 The use of protease improves the growth performance of newly weaned piglets fed diets reduced in energy and protein.** O.O Babatunde*, G Tactacan, M.S Vieira, L Lahaye, and M.L de Moraes, *Jefo Nutrition Inc., St-Hyacinthe, QC, Canada*. Optimizing protein and amino acid utilization in nursery pig diets enhances feed efficiency and supports sustainable agriculture by reducing nitrogen loss and improving intestinal health. This study aimed to evaluate the effects of protease supplementation on the growth performance of newly weaned nursey pigs. A total of 288 weaned piglets (24 days old, initial body weight (BW) = 6.9 ± 0.03 kg) were randomly assigned to one of 3 dietary treatments for 21 days. Treatments included a negative control (NC) diet reduced in crude protein (-5%), digestible lysine (-5%), and energy (-15 kcal/kg net energy) relative to dietary requirements, and the NC supplemented with protease at 125 or 250 g/t. Each dietary treatment had eight replicates with 12 pigs per pen, balanced by gender. Growth performance parameters including final BW (FBW), average daily feed intake (ADFI), average daily gain (ADG), and feed conversion ratio (FCR) were measured at days 14 and 21. Data were analyzed using ANOVA with significance set at $P$ < 0.05. Treatment differences were determined using linear contrast and Tukey's HSD test. At 14 days, there was no difference in the ADFI of pigs in all treatments, however, supplementing the highest dose of protease significantly increased ($P$ < 0.05) the FBW and ADG of pigs compared to the NC. Additionally, there was a linear improvement ($P$ < 0.05) in the FCR of pigs with increasing levels of protease. At day 21, there was no difference in ADFI but a linear improvement ($P$ < 0.05) in FCR with pigs fed NC supplemented with protease at 250 g/t having a 7% improvement in feed conversion as compared with pigs fed the NC. In conclusion, protease supplementation in protein- and energy-reduced nursery pig diets improved piglet growth and feed efficiency, with the high dose of 250 g/t outperforming the standard 125 g/t. The linear improvement in performance highlights enhanced nutrient utilization, promoting efficient and sustainable swine production. **Key Words:** growth performance, piglets, protease

## Theme II Posters: Advances in Understanding of Nutrient Digestion and Absorption

**127 Pancreatic enzyme activity and intestinal morphology in pigs with low and high feed conversion ratios fed three different levels of crude protein.** J.M. van der Linden[1], M.E. van der Heide[2], M. Barszcz[3], A. Konopka[3], A. Tusnio[4], E. Swiech[4], K Gawin[4], J.V. Nørgaard[2], and J.G. Madsen*[1], [1]*Department of Veterinary and Animal Sciences, Faculty of Health and Medical Sciences, University of Copenhagen, 1870 Frederiksberg, Denmark*, [2]*Department of Animal and Veterinary Sciences, Faculty of Technical Sciences, Aarhus of University, 8830 Tjele, Denmark*, [3]*Laboratory of Analysis of Gastrointestinal Tract Protective Barrier, Department of Animal Nutrition, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Instytucka 3, 05-110 Jablonna, Poland*, [4]*Department of Animal Nutrition, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Instytucka 3, 05-110 Jablonna, Poland*. Reducing the feed conversion ratio (FCR) in pigs during the grower-finisher phase is crucial for improving the herd's economy, while also reducing nutrient excretion and carbon footprint. Therefore, understanding the biological factors contributing to FCR is important. In this study, sixty grower-finisher gilts were divided between three dietary treatments and fed a low (LCP; 148 g CP/kg), standard (SCP; 152 g CP/kg), or high (HCP; 158 g CP/kg) crude protein (CP) diet from 30 to 115 kg of body weight, for a period of 10 weeks. Feed intake and weight were measured individually. Within each diet, pigs were categorized based on their FCR during the final four growing weeks as the 30% highest (high FCR), medium 40%, and 30% lowest (low FCR). The pigs categorized as low and high FCR were euthanized at 20 weeks of age where pancreatic as well as small (SI) and large intestinal samples were collected, and subsequently subjected to an analysis of enzyme activity and morphological characteristics. The data were analyzed in R using a mixed linear model applying ANOVA to evaluate the fixed effect of FCR category, dietary treatment and their interaction on pancreatic enzyme activity and morphological parameters of the small and large intestine. Differences with respect to enzyme activity in low compared with high FCR pigs included lower ($P$ = 0.05) trypsin activity, and greater ($P$ = 0.02) alanine aminotransferase activity. There was a tendency for a diet × FCR interaction with respect to aspartate aminotransferase ($P$ = 0.06), and a tendency ($P$ = 0.08) for a FCR effect on lactate dehydrogenase activity. In addition, differences in gut morphology between low and high FCR pigs were found, where a thinner ($P$ = 0.04) caecal muscular layer and a tendency ($P$ = 0.07) for shorter distal SI villous height were observed in the low FCR pigs. In conclusion, the results indicate a possible link between the level of feed efficiency of grower-finisher gilts and their pancreatic enzyme activity. **Key Words:** Grower-finisher pigs, feed efficiency, pancreatic enzyme activity

**128 The role of alpha-amylase, in comparison to mixed pancreatic enzymes, for the maintenance of small intestinal wall structure in a model of exocrine pancreas insufficiency in pigs.** Kamil Zaworski*[1], Dominika Szkopek[2], Jaroslaw Wolinski[2], Stefan Pierzynowski[3,4], Kateryna Pierzynowska[1,3], and Björn Weström[3], [1]*Department of Animal Physiology, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jablonna, Poland*, [2]*Large Animal Models Laboratory, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jablonna, Poland*, [3]*Department of Biology, Lund University, Lund, Sweden*, [4]*Department of Medical Biology, Lublin, Poland*.
The study aimed to compare the effects of porcine pancreatic enzymes (Creon®) and microbial-derived alpha amylase (MD amylase) on small intestine structure, mucosal glycogen accumulation, and enterocyte turnover in pigs with surgically induced exocrine pancreatic insufficiency (EPI). Eighteen pigs (initial BW 15.0 ± 2.3 kg) were divided into three groups: EPI (n = 6, no supplementation), Amylase (n = 6, MD amylase at 2 × 4,000 units/day), and Creon (n = 6, Creon® at 2 × 100,000 units/day). Four healthy pigs served as controls. Enzymes were administered for 30 days in a 54-day study. Histomorphometric and immunohistochemical analyses assessed crypt stem cell proliferation, epithelial apoptosis, and mucosal glycogen levels. Data were analyzed using ANOVA and Kruskal-Wallis tests, with normality checked via Shapiro-Wilk test. EPI caused small intestine mucosal atrophy, reducing villi length, crypt depth, and mucosal/muscularis thickness

compared to controls ($P < 0.05$). Enzyme supplementation reversed these changes ($P < 0.05$). EPI increased apoptotic and decreased proliferative activity, both restored by Creon® and MD amylase. Glycogen accumulation in the duodenum rose significantly in EPI pigs (710.6 ± 317.4 µg/mg vs. 48.8 ± 21.1 µg/mg in controls) but was unaffected by supplementation. In conclusion, pancreatic enzyme supplementation improves intestinal morphology and cellular activity in EPI, with MD amylase showing effects comparable to Creon®. **Key Words:** alpha amylase; pancreatic enzymes; exocrine pancreatic insufficiency;

**129 The postprandial absorption of protein as peptides (di-, tripeptides) and free amino acids in exocrine pancreas insufficient (EPI) pigs.** Kamil Zaworski*[1], Kateryna Pierzynowska[1,2], Wieslaw Szczesny[3], Stefan Pierzynowski[2], and Piotr Wychowanski[4,5], [1]*Department of Animal Physiology, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jablonna, Poland*, [2]*Department of Biology, Lund University, Lund, Sweden*, [3]*Institute of Information Technology, Warsaw University of Life Sciences, Warsaw, Poland*, [4]*Department of Head and Neck and Sensory Organs, Division of Oral Surgery and Implantology, Institute of Clinical Dentistry, Gemelli Foundation for the University Policlinic, Catholic University of the "Sacred Heart", Rome, Italy*, [5]*Departament of Interventional Dentistry, Collegium Medicum, Nicolaus Copernicus University, Bydgoszcz, Poland*. The impact of exogenous digestive enzymes supplementation on dietary protein digestion and absorption of nutritional peptide relative to amino acids was tested with varying mixed meal tolerance tests (MMTT) on eighteen, male pigs before and after exocrine pancreatic insufficiency (EPI) development using adapted ninhydrin method. The study used 3 groups of animals: the Control group (n = 6) was fed only, Creon group (n=6) was fed + Creon® (2 x 100,000 units per day), Amylase group was fed + Amylase (2 x 4,000 units per day). The duration of the experiment was 54 days, the first MMTT was performed in healthy animals, the second after the development of EPI. The quantitative amine group estimation using the modified ninhydrin method was combined with an algorithm developed to allow for estimation of the composition of di- and tripeptides as well as free amino acids of dietary origin in in blood. The absorption of free amino acids from dietary protein varies from between 15 to 20%, with the rest of the amino acids from dietary protein absorbed in the form of dipeptides (20 – 30 %) and tripeptides (30 – 65%), as estimated by the algorithm we developed. Creon, as well as Amylase treatment, increased the absorption of amino acids in the form of tripeptides, in both healthy and EPI pigs $P > 0.05$. Moreover, both Creon and Amylase increased the total absorption of amino acids in EPI pigs, as represented by amine group equivalents $P > 0.05$. In summary, a modified ninhydrin method and supporting algorithm allow with good probability the ability to estimate the ratio and rate of postprandial di-, tripeptide and free amino acids absorption from dietary protein. Adaptation to the diet macronutrients (protein) as well as digestive enzymes additives improve absorption of protein in the form of di-, and tripeptides. **Key Words:** protein digestion; amino acids; peptide;

**130 Low Protein Diet Enhances Plasma Abundance of Pantothenate in Lactating Sows Under Heat Stress.** Astrid Coba*[1], Xinle Tan[1], Maximiliano Muller[1], Elham Assadi Soumeh[2], Marta Navarro[1], and Eugeni Roura[1], [1]*Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Brisbane, Queensland, Australia*, [2]*School of Agriculture and Food Sustainability, The University of Queensland, Brisbane, Queensland, Australia*. As climate change intensifies heat stress (HS) episodes, high performing lactating sows (and their litters) are increasingly vulnerable due to high metabolic demands and reduced feed intake under HS. While physiological responses like increased respiration rate (RR) aid cooling, these mechanisms often fail to prevent performance decline. Previous studies from our group demonstrated that a low crude protein diet (LP) significantly reduced RR in sows under HS conditions, indicating its potential to support heat resilience. This study evaluated differences on the plasma metabolome of lactating sows under HS when fed a LP compared to standard crude protein (SP) diet. It was hypothesised that LP compared to SP enhances thermoregulatory resilience by optimizing metabolic pathways. Fifteen Large White sows (308±7.84 kg, parity 4.53±0.49) underwent a crossover thermal protocol alternating

between thermoneutral (TN, 20°C) and HS (27°C) conditions, with 60% relative humidity. Sows were fed either a SP (19% CP, 1% SID Lysine) or LP (16% CP, 1% SID Lysine) diets. Plasma samples were collected on day 3 of each thermal period. Untargeted LC-MS analysis identified 36 differentially abundant metabolites (DAM). Statistical analysis used a linear mixed model with "Period," "Room," "Diet," "Environment," and their interactions as fixed effects, and "Sow" and "Sow×Period" as random effects, followed by a Tukey post hoc test in RStudio. Significance was set at $P \leq 0.05$. The analysis revealed that under HS, the LP diet significantly increased pantothenate levels ($P < 0.05$), suggesting enhanced lipid utilisation—a more heat-efficient metabolic pathway. Conversely, nicotinamide levels were higher in the SP group ($P < 0.05$), indicating reliance on glycolytic and oxidative pathways, which are associated with greater heat production. In conclusion, reducing dietary CP (LP) improved thermoregulatory resilience supporting sustainable production under HS associated with increased plasma pantothenate thus indicating increased lipid metabolism. **Key Words:** Heat stress, Low crude protein diet, Lactating sows

**131 Lys or Thr deficiencies reduced growth rates associated with decreasing tail biting in piglets while tail-biters showed signs of disrupted amino acid metabolism in plasma.** A. Abdallah*[1], A. Kumar[1], M. Navarro[1], M. Muller[1], X. Tan[1], A. Tilbrook[1,2], K. J. Plush[3], D. N. D'Souza[3], and E. Roura[1], [1]*Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, St Lucia, QLD 4072, Australia*, [2]*School of Veterinary Science, The University of Queensland, Gatton, QLD 4343, Australia*, [3]*SunPork Group, Eagle Farm, QLD 4009, Australia*. Dietary amino acid (AA) deficiencies have been signalled as a potential risk factor for tail biting (TB) in pigs. This study aimed to identify metabolic biomarkers associated with TB. We tested two hypotheses: (i) that AA deficiencies increase TB incidence**;** (ii) tail-biter (perpetrator -PP-) identified by direct observation across all pens have elevated plasma metabolites indicative of increased AA turnover compared to non-perpetrator (NP) pigs. A total of 432 piglets (LW X LD, 27±3 days of age) with intact tails were allocated to four dietary treatments: control or 35% deficiency in Lys, Met, or Thr. A randomized complete block design with 24 pens of 18 pigs (50% entire males) with a wide range of body weights within each pen was used. Diets were isonitrogenous and isoenergetic, using Ala to substitute the deficient AAs. After five weeks, blood samples were collected from one NP per pen (n = 24) and all PP (n = 15). The effect of diet on TB was analysed using the GLM procedure of SAS with diet and block as fixed effects and pen as the unit. Plasma metabolites were analysed using mass spectrometry (SWATH-MS). Partial Least Squares-Discriminant Analysis (PLS-DA) was performed using MetaboAnalyst 6.0. VIP scores ≥ 1.5 were used to select metabolites based on the differentiation between PP and NP with pig as the unit. Statistical significance was determined using unpaired Wilcoxon rank-sum test with a false discovery rate of $P < 0.01$ and a fold change threshold of 2. The AA treatments decreased ($P < 0.05$) accumulated TB incidence to 48.1% and 42.6% for Lys, and Thr, respectively, compared to 82.4% in the control. These results were associated with reduced weight gain (hence lower animal density) in the AA-deficient groups which likely impacted TB incidence. When comparing PP to NP, twelve metabolites were elevated in the PP group, four including O-Acetylserine, glutamic acid, valine, and betaine, were indicators of increased AA turnover which may explain TB behaviour in PP pigs. **Key Words:** Tail biting, amino acids, metabolomics

**132 Protein fermentation biomarkers do not vary between dietary protein sources differing in digestibility.** R. Minderhoud*[1,2], E. Capuano[2], S. de Vries[3], A. Even[4,5], and G. Hooiveld[1], [1]*Division of Human Nutrition and Health, Wageningen University & Research, Wageningen, The Netherlands*, [2]*Food Quality and Design Group, Wageningen University & Research, Wageningen, The Netherlands*, [3]*Animal Nutrition Group, Wageningen University & Research, Wageningen, The Netherlands*, [4]*OnePlanet Research Center, Wageningen, The Netherlands*, [5]*imec, Wageningen, The Netherlands*. Microbial fermentation of undigested proteins in the large intestine may result in the production of metabolites that are potentially harmful to gut epithelia and host health. While increased protein intake is known to increase the flow of proteins into the large intestine, and in turn, of protein fermentation, the effect of protein digestibility on this process is unknown. To investigate this,

a fully controlled randomized cross-over human dietary intervention study with 2 protein sources that differed in digestibility was performed in 15 participants. For 7 days participants consumed a diet supplemented with 30 grams of protein per day (divided over 3 meals) either from Bovine Plasma Protein, as low digestible protein (LDP), or Whey Protein Isolate, as high digestible protein (HDP) source, with a 7-day washout period in between. Macronutrient content was identical for both diets with 12% proteins, 36% lipids, 50% carbohydrates, and 2% fiber (w/w) of total energy intake. The diet contained acid-insoluble-ash (AIA) as indigestible marker to quantify fecal biomarkers. Pooled fecal and morning urine samples from day 4 onwards were used for analysis. No differences between diets were found for fecal ammonia (mean ± SD: LDP 16 ± 6 vs HDP 17 ± 5 mg/g AIA, $P$ = 0.743), SCFA (LDP 1807 ± 1085 vs HDP 1987 ± 1575 µmol/g AIA), and BCFA (LDP 134 ± 52 vs HDP 139 ± 43 µmol/g AIA) concentrations ($P$ > 0.05), and urea in urine (LDP 19 ± 5 vs HDP 22 ± 3 mmol/mmol creatinine, $P$ = 0.595), as determined by linear mixed-model analysis with participant as random effect (n = 13). In conclusion, protein fermentation biomarkers measured in feces and urine were not increased by consumption of a LDP source compared to a HDP source. High individual variation between participants suggests that individual differences strongly influence metabolite kinetics. Insights from human protein digestibility and fermentation also hold relevance for understanding pig digestive processes. **Key Words:** protein fermentation, protein digestibility, biomarkers

**133 Peri-natal conditioning and early feed intake in piglets.** E. Roura*, S. Taylor, and M. Navarro, *Queensland Alliance for Agriculture and Food Innovation (QAAFI), The University of Queensland, St Lucia, Australia*. Early weaning practices are associated with disruptive social and environmental challenges leading to the post-weaning stress syndrome in piglets characterised in part by a reduced nutrient intake during the transition between sow milk and solid feed. This post-weaning anorexia is fueled by a neophobic event understood to be driven by taste and olfactory cues. Peri-natal conditioning relevant to feed intake relies on sensory cues triggered by volatile organic compounds (VOCs) forming olfactory cues familiarizing piglets with feed ingredients present in maternal diets during foetal development and early after birth. We have coined the concept of “iso-sensorial diets” to refer to two feeds formulated to meet different nutrient requirements (e.g., gestating compared to post-weaning diets) while maintaining a continuity in the primary olfactory profile based on the VOCs profile. Firstly, formulating iso-sensorial diets required a complete characterisation of the volatilome of the main ingredients used in sow feeds. The volatilome of main cereals (e.g., wheat and barley), pulses and protein sources (e.g., canola, soybean meal, and meat and bone meals) were determined by HES-SPME/GC-MS, resulting in more than 600 VOCs identified belonging to a variety of chemical groups. The transfer rates of the VOCs from sow feed to amniotic and other maternal fluids revealed a few key volatiles suitable to depict the iso-sensorial diet concept. These included hexanal, ethyl hexanoate, 1-octen-3-ol, pentanol, penten-3-ol, and α-pinene, among others. Formulating post-weaning diets with an iso-sensorial principle based on gestating diets resulted in reduced post-weaning stress indicated by improved feed intake and reduced diarrhoea compared to standard hetero-sensorial practices in piglets. In summary, maternal conditioning to VOCs and the application of iso-sensorial diet formulation principles have the potential to improve the health and welfare of post-weaning piglets. **Key Words:** weaning anorexia volatinome

**134 Identification of metabolic events responsive to oxidized soybean oil in the luminal metabolome of nursery pigs.** Q. Mao[1], J. Yuan[1], B. J. Kerr[2], and C. Chen*[1,3], [1]*Department of Food Science and Nutrition, University of Minnesota, St. Paul, MN, USA*, [2]*USDA-ARS National Laboratory for Agriculture and the Environment, Ames, IA, USA*, [3]*Department of Animal Science, University of Minnesota, St. Paul, MN, USA*. The negative influences of feeding thermally oxidized oils on metabolic health and growth performance of pigs start with the introduction of their lipid oxidation products (LOPs) into the gastrointestinal tract (GIT). However, the changes in luminal metabolome after feeding and their correlations with animal performance have not been examined previously. In this study, 60 weaning barrows (3 pigs/pen, initial average BW of 6.2 kg)

were fed the nursey diets containing control soybean oil (CSO) or heated soybean oil (HSO) for 28 days for performance, and then 10 focal pigs per treatment (1 pig/pen) were harvested for luminal digesta from stomach, duodenum, proximal jejunum, ileum, cecum, and colon, as well as liver samples. The metabolite profiles of luminal digest and liver samples, together with the chemical composition of HSO and CSO, were determined by the liquid chromatography-mass spectrometry-based metabolomic analysis. The results on the concentrations and the relative abundance of metabolites, and enzyme activities were analyzed by unpaired t-test to compare CSO and HSO groups with a *P*-value < 0.05 as significant and 0.05 < *P*-value < 0.1 as a trend. HSO feeding decreased weight gain and feed intake while increased liver/body weight ratio. The analysis of the luminal metabolome showed that HSO feeding, site-specifically, increased the aldehydes and protein carbonylation in digesta, due to continuous disposition and *de novo* production of aldehydes along the GIT. HSO suppressed digestive activities as shown by the decreases of free amino acids and elastase activity in the small intestine and the decreases of bile acids in both GIT and gallbladder. Many HSO-elicited changes in the luminal metabolome had site- and chemical-specific correlations with growth performance and the metabolic changes in the liver, indicating their potential causative functions in HSO-induced adverse effects and their potential values as the targets of mitigation treatments. **Key Words:** oxidized lipids, luminal metabolome, metabolic markers

**135 Nutrient and energy digestibilities of defatted corn germ in pigs of different ages.** H Moreira Junior*[1], J. A. L Barbosa[1], J. L. Brito[1], C. E. M. Bertanha[1], S. S. S. Souza[1], A. Gorrosterrazú[1], A. B. S. Oliveira[2], M. L. P. Tse[3], and U. S. Ruiz[1], [1]*University of São Paulo (USP), Luiz de Queiroz College of Agriculture, Department of Animal Science, Piracicaba, SP, Brazil, 13418-900*, [2]*Ingredion, Mogi Guaçu, SP, Brazil, 13841-010*, [3]*São Paulo State University (UNESP), School of Veterinary Medicine and Animal Science, Department of Animal Production, Botucatu, SP, Brazil, 18618-970*. The capacity of pigs to digest nutrients and energy of feed ingredients may vary with age, however, the nutrient composition of ingredients is fixed in most feed composition tables. Thus, the objective of this study was to investigate the nutrient digestibility of defatted corn germ (DCG) in female pigs at four phases: 28 days old (7.81 ± 0.68 kg body weight - BW), 78 days (34.23 ± 3.42 kg BW), 125 days (79.79 ± 4.79 kg BW), and 243 days (169.00 ± 6.51 kg BW). The standardized ileal digestibility (SID) of amino acids (AA) and crude protein (CP), the apparent total tract digestibility (ATTD) of dry matter (DM), neutral detergent fiber (NDF), gross energy (GE), and digestible energy (DE) of DCG were analyzed. Forty-eight female pigs, 12 pigs per phase, were fed a nitrogen-free (NF) diet, to determine endogenous AA losses, or a diet containing 40% DCG, replacing starch, sugar, cellulose, and oil in NF diet, as the sole source of AA and protein. Both diets contained 0.5% titanium dioxide, used as an indigestible marker. The animals were fed at 2.8 x maintenance DE for 14 days (12 days adaptation and 2 days feces collection). On the 15th day, the pigs were euthanized to ileal digesta collection. A randomized block design was adopted, with 4 treatments (pig ages), 6 replicates per treatment, and 1 animal per experimental unit. Data were subjected to analysis of variance using the MIXED procedure of SAS and to regression analysis. The digestibility of energy and nutrients of DCG linearly increased in female pigs at the initial phase to the adult sows. The enhancements ($P < 0.05$) in the ATTD of nutrients and energy were as follows: DM, 71% (42.8 to 73.3%); NDF, 24% (65.4 to 80.8%); GE, 112% (37.1 to 78.5%). The DE increased ($P < 0.05$) from 1849 kcal/kg to 3914 kcal/kg and the SID of total AA and CP augmented ($P < 0,05$) from 54.4% to 90.7%, and from 52.5 to 84.2%, respectively. In conclusion, the age of the pigs has an important effect on nutrient and energy digestibility of DCG. **Key Words:** Ileal Digestibility, Age of Pigs, Co-products

**136 Evaluation of Soluble Monosaccharides as a Proxy for Estimating Mucin Protein Concentration in Ileum Digesta.** I. Kaikat[1], S. Tibble*[2], L. Blavi[2], M. A. Ton Nu[2], A. Koppenol[2], G. González-Ortiz[3], A. Acosta-Lagaxio[1], K. Englyst[4], and J. F. Pérez[1], [1]*Animal Nutrition and Welfare Service (SNiBA), Department of Animal and Food Science, Universitat Autònoma de Barcelona (UAB), 08193 Bellaterra, Spain*, [2]*AB Neo, PL Fraga, C/ Comunidad de Murcia, parc.*

*LIE-1-03, 22520 Fraga (Huesca), Spain,* [3]*AB Vista, Marlborough SN8 4AN, United Kingdom,* [4]*Englyst Carbohydrates Ltd, 2 Venture Road, Southampton Science Park, Southampton SO16 7NP, UK.* This study aimed to analyze soluble monosaccharides (fucose, galactose, and N-acetylglucosamine [GlcNAc]) in ileal and proximal colon digesta of early-weaned piglets to evaluate the effects of cereal type on mucin secretion and its link to endogenous nitrogen excretion. Mucin, a heavily glycosylated protein, is the primary component of gastrointestinal mucus and contributes significantly to endogenous losses in the ileum. Forty-eight piglets (21d old, 5.5±0.8 kg) were allocated to 16 pens (4 pens/diet; 3 pigs/pen) and fed diets containing 92% maize, wheat, barley, or extruded barley, with sucrose and casein. Ytterbium oxide (50mg/kg) was included as an inert marker to estimate nutrient flows. On day 15, ileal and proximal colon digesta samples were collected, and nutrient flows were calculated per kg of cereal consumed. Cereal composition influenced monosaccharide profiles. Fucose and GlcNAc contents were absent in all cereals, while soluble galactose content ranged from 0.8g/kg in maize to 1.75g/kg in wheat. In the ileum, flows ranged from 0.44 to 1.81g/kg for soluble fucose, 1.74 to 6.81g/kg for soluble galactose, and 1.31 to 3.55g/kg for soluble GlcNAc, with barley and wheat producing the highest flows. Proximal colon flows were lower, ranging from 0.25 to 0.6g/kg for fucose, 0.61 to 1.76g/kg for galactose, and 0 to 0.63g/kg for GlcNAc. These reductions suggest that 60–80% of mucins were fermented in the caecum and proximal colon. Positive correlations among monosaccharides ($r$ = 0.962 for s.GlcNAc/s.fucose, $P$ < 0.001; $r$ = 0.773 for s.galactose/s.fucose, $P$ = 0.005) indicate their endogenous and/or microbial origins. These results suggest that differences in cereal composition, particularly fiber fermentability, influence mucin secretion and fermentation. Soluble monosaccharides may serve as proxies for estimating mucin secretion, offering a tool to refine ileal digestibility predictions in vitro. **Key Words:** Mucin secretion, Monosaccharide fermentation, Piglets

**137 Testing a new index of dietary nitrogen to study piglet performance and gut health.** F.A. Eugenio, N. Vieco-Saiz, J. Consuegra*, T. Mahmood, and Y. Mercier, *Adisseo France S.A.S, Saint-Fons, France*. Reducing crude protein (CP) of piglet diets is a common strategy to limit unabsorbed and excess nitrogen (N) that can be fermented in the hindgut, potentially harming gut health and performance. However, results of this strategy are inconsistent likely because CP is an imperfect designation of feed N. We hypothesize that non-retained nitrogenous compounds (NRNC), calculated as CP minus the sum of digestible amino acids (dAA), is a better indicator of the negative effects of excess N on performance, intestinal health, and diarrhea scores in piglets than CP. Ingredients with high (i.e., animal sources) and low (i.e., plant sources) NRNC levels were identified based on the sum of table values of dAA (all 20) subtracted from CP, and were used to formulate our diets. We used 192 weaned piglets (8.38 ± 1.47 kg), blocked by initial weight and allocated in pairs to 96 pens. Each pen was randomly allotted to one of 8 diets in a full-factorial arrangement with 4 levels of NRNC (2.5, 4, 5.5, and 7%) and 2 levels of CP (17.5 and 20%). The pigs were fed these diets *ad libitum* for 28 days. Body weight, feed intake, and daily diarrhea score of each pen were recorded. Data were analyzed using two-way ANOVA and GLMMIXED with NRNC, CP, and their interaction as main factors, and block as a random factor for zootechnical data and diarrhea scores, respectively. No interactions were observed ($P$ > 0.10). Gain to feed was significantly ($P$ = 0.01) inversely related with NRNC levels, while CP only tended to influence this parameter ($P$ = 0.06). Other zootechnical parameters were unaffected by either factor ($P$ > 0.10). Diarrhea score was higher in piglets fed higher CP ($P$ = 0.01) but was not influenced by NRNC ($P$ = 0.27). In conclusion, our results showed that NRNC appears to a good predictor of piglet performance while CP was a better predictor of diarrhea incidence. These findings highlight the importance of considering dietary N sources beyond CP to optimize piglet health and performance. **Key Words:** dietary protein, intestinal health, piglet

**138 Evaluating Silicon as an Alternative Indigestible Marker for Dry Matter Digestibility in Swine.** I. Kaikat*[1], E. Llauradó-Calero[1], A. Cerisuelo[2], D. Torrallardona[3], and D. Solà-Oriol[1], [1]*Animal Nutrition and Welfare Service (SNiBA), Department of Animal and Food Science, Universitat Autònoma de Barcelona (UAB), 08193 Bellaterra, Spain,* [2]*Centro de*

*Investigación y Tecnología Animal (CITA), Instituto Valenciano de Investigaciones Agrarias (IVIA), 12400 Segorbe, Spain,* [3]*Animal Nutrition, Institute of Agrifood Research and Technology (IRTA), 43120 Constantí, Spain*. This study aimed to evaluate silicon (Si), an endogenous element, as a marker for estimating dry matter digestibility (DMD) in swine compared to added markers, such as acid-insoluble ash (AIA), titanium dioxide (TiO2), and ytterbium oxide (Yb2O3) across four trials (T). T1 tested four diets with varying protein sources in pigs (Yb2O3 0.005% and Celite 1%), T2 compared standard and high-fiber diets in piglets (TiO2 0.5% and Celite 1%), and T3 (Celite 1%) and T4 (TiO2 0.5%) evaluated diets differing in phytase doses in lactating sows. Ti, Yb, and Si concentrations were determined in diets and feces using ICP-OES 5900, and AIA was analysed following ECR nº152/2009. The inherent Si concentrations (1.8-2.5g/kg in Celite-supplemented diets and 0.25-0.30g/kg in non-supplemented) were used to calculate DMD, alongside added markers. Statistical analyses included diet, marker, and their interaction as fixed effects. No significant interactions between diet and marker were observed across all trials. In T1, AIA, Yb, and Si provided similar DMD estimates (84.6, 84.7, and 84.5%, respectively; $P = 0.708$) with strong correlations between Si and AIA ($r = 0.74$) and Si and Yb ($r = 0.64$). In T2, Ti and Si yielded consistent results with no significant differences (80.2 and 80.9%, respectively; $P = 0.081$; $r = 0.85$). In T3 and 4, Si provided lower DMD values compared to AIA (78.8 vs. 82.3%; $P < 0.001$) and Ti (80.3 vs. 84.9%; $P < 0.001$), while still consistently estimating differences between diets with good correlations (Si vs. AIA, $r = 0.65$; Si vs. Ti, $r = 0.51$). Si shows promise as a digestibility marker under commercial conditions. Its endogenous presence eliminates the need for supplementation, reducing cost and simplifying diet formulation. Si's integration with routine mineral analyses and applicability to both trials and on-farm diets enhances its practicality and versatility. Future research should focus on optimizing Si as a reliable marker. **Key Words:** Feed evaluation, Endogenous marker, Nutrient digestibility

**139 Unveiling the impact of dietary net energy reduction on nutrients partition patterns of growing pig: A modeling approach.** W. Ren*[1], J. C. Zhang[1], Z. Z. Wang[1], S. K. Wang[1], A. J. Cowieson[2], H. X. Zhai[1], and E. Perez-Calvo[2], [1]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Bazhou, China*, [2]*dsm-firmenich, Animal Nutrition and Health, Kaiseraugst, Switzerland*, [3]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Tulln, Austria*. Adjusting dietary net energy (NE) density in response to fluctuations in high-energy ingredient prices is essential to pig producers. But the extent to which reduced NE density impacts growth performance remains unclear. This study investigated the effects of dietary NE reduction on growth performance, apparent total tract digestibility (ATTD) and blood profiles, with the aim of developing an energy deficiencies model. Fifty-five growing pigs (39.44 ± 3.79 kg) were allocated to 5 dietary treatments over a 35-day period. Pigs were housed by treatments and fed ad libitum by automatic feeding system. Treatment A met the nutritional requirements, while treatments B to E represented a controlled reduction in NE by -47, -95, -142 and -189 kcal/kg, respectively. Growth performance was recorded individually by automatic feeding system. Feces and blood samples were collected at d 35 to analyze ATTD and blood profile indicators, including total protein, albumin, globulin, and blood urea nitrogen (BUN). The results were analyzed by JMP and the significance was defined at $P < 0.05$. No statistical differences were detected between treatments on growth performance but pigs in treatment D and E showed numerically lower growth performance. A significant quadratic response in ATTD of dry matter, crude protein and gross energy were observed. The ATTD of DM (85.98 *vs.* 90.13%), CP (79.57 *vs.* 85.65%), and GE (85.35 *vs.* 89.80%) in treatment E were significantly lower than treatment A, which was associated with the numerically reduced growth performance. Moreover, BUN concentrations showed quadratically response as NE density reduced. Compared to A, treatment D and E showed significantly lower BUN concentration (11.18 and 11.36 vs. 16.64 mg/L), aligning with expectations for animals impaired protein deposition. In conclusion, NE gradient reduction until 189 kcal/kg in grower pigs had limited impact on growth performance but significantly impaired nutrient digestibility and BUN levels. **Key Words:** Net energy reduction, Growth performance, Nutrient digestibility

**140 Kinetics of in vitro protein solubilization of diets including various protein sources is affected by grinding and pelleting.** S Zhang[1,2], L de Jonge[1], S de Vries[1], V Lagos[3], F Molist[3], and W.J.J. Gerrits*[1], [1]*Animal Nutrition Group, Wageningen University & Research, ,Wageningen, The Netherlands*, [2]*State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China*, [3]*Schothorst Feed Research, Lelystad, The Netherlands*. This study aimed to quantify the effects of grinding, pelleting, and their interaction on rate and extent of *in vitro* nitrogen (N) solubilization in a 2×2 factorial arrangement of treatments across five diets. Wheat, barley, faba beans (FB), sunflower seed meal (SFM), and rapeseed meal (RSM) were coarsely (D50 = 587-1618 µm) or finely (D50 = 245-304 µm) ground and mixed with a maize-soybean basal diet (50:50) in a 60:40 ratio. Diets were either pelleted (80°C, 20 kWh/metric ton) or in mash form, creating 20 test diets, with two additional basal diets (mash fine, pellet fine) included. Samples were incubated (2 replicates) with pepsin at pH 4 for 90 min, then at pH 2 for 90 min, followed by small intestine simulation at pH 6.8 for 15 min, then with pancreatin added for 240min. Solubilized N was measured at several time points (0-435min). The rate of N solubilization varied more among treatments during the stomach phase than the small intestine phase. Fine wheat, barley, and FB diets showed a higher rate of N solubilization during 0-90min of the stomach phase at pH 4 compared with their coarse diets ($P < 0.05$). By the end of the small intestine phase, fine barley and FB diets had ~7% higher N solubility than coarse diets. Pelleting increased the rate of N solubilization for wheat, barley, FB, and RSM diets during 0-90min of the stomach phase but decreased it for SFM diets ($P < 0.05$). For wheat, reduced particle size and thermal treatment from pelleting increased the rate of N solubilization during 0-90min of the stomach phase for the pellet diets compared with the mash diets (interaction, $P < 0.05$). By the end of the small intestine phase, pellet barley and FB diets had ~8% higher N solubility than mash diets. Overall, the effects of grinding and pelleting on the kinetics of N solubilization were more pronounced in the diets including native ingredients compared with the diets including pre-processed ingredients. **Key Words:** Processing, Particle size, Digestion kinetics

**141 In vitro starch degradation kinetics of diets containing different starch-rich ingredients is affected by ingredient particle size and energy degree input pelleting.** S Zhang[1,2], V Lagos[3], L de Jonge[1], S de Vries[1], W.J.J. Gerrits[1], and F Molist*[3], [1]*Animal Nutrition Group, Wageningen University & Research, Wageningen, The Netherlands*, [2]*State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China*, [3]*Schothorst Feed Research, Lelystad, The Netherlands*. Feed technology plays a crucial role in regulating kinetics of starch digestion in pig diets. The study aimed to quantify the effect of ingredient grinding, feed pelleting conditions, and their interaction on the rate and extent of in vitro starch degradation in a 2×2 factorial arrangement of treatments across various feeds. Wheat, barley, maize, and faba beans (FB) were coarsely (D50 = 1348-1618 µm) or finely (D50 = 245-343 µm) ground, and mixed with a finely (D50 = 366 µm) ground basal diet (BD; 50% maize, 50% soybean meal) in a ratio of 60:40. Subsequently, diets were pelleted applying different combinations of specific thermal energy (STE; 60 or 80°C in the conditioner) and specific mechanical energy (SME; low = 10 or high = 20 kWh/metric ton in the expander). The test diets and BD were pelleted applying either a high STE/high SME (80H) or a low STE/low SME (60L) process, resulting in four treatments (coarse 60L, coarse 80H, fine 60L, fine 80H) per diet. Starch digestion kinetics of all pelleted test feeds and BD (fine 60L, fine 80H) were determined in duplicate via in vitro hydrolysis by pancreatin, amyloglucosidase, and invertase. Aliquot of suspensions were collected at 0, 30, 60, 90, 120, and 240 min. Results indicated that fine grinding increased the extent of starch degradation for all diets (grinding effect; $P < 0.05$). Higher rates of starch degradation were observed in all 80H compared with 60L diets (pelleting effect; $P < 0.05$), but the effect was more pronounced in the FB diets. The enhancement of starch degradation in fine-ground diets was amplified under high-energy pelleting conditions (interaction; $P < 0.01$), particularly in wheat diets, demonstrating a synergistic effect. Overall, the effects of particle size and pelleting conditions on the kinetics of starch degradation strongly depend on the starch origin and may not be additive. The results can

be used to tailor processing conditions and choice of ingredients towards a targeted starch digestion kinetics. **Key Words:** Processing, grinding fineness, digestion kinetics

**142 Determining NDF fermentability using an *in vitro* fermentation model to estimate *in vivo* apparent total tract fermentability in growing-finishing pigs.** Qiong Hu*, Patricia Pluk, and Sandra Paredes, *Cargill Animal Nutrition and Health, Minneapolis, MN*. The role of fiber in swine nutrition has become crucial due to its health and performance benefits. Hence, accurate determinations of neutral detergent fiber (NDF) apparent total tract fermentability (ATTF) and its change through a pig's life are relevant. It was hypothesized that *in vitro* fermentation would correlate with pig ATTF on common ingredients and ATTF would increase with age. A total of 128 growing-finishing (GF) pigs (n=8, initial body weight=22.1±1.7 kg) blocked by weight and sex were randomly allotted to 1 of 16 treatments (1 control and 15 ingredients - wheat bran, red dog, wheat middling, wheat, sunflower hulls, sunflower meal, soybean hulls, soybean meal, rice hulls, high-fat rice bran, defatted rice bran, corn, DDGs, barley, rapeseed meal) in a completely randomized block design over 4 phases. Experimental diets were corn soy wheat based. The control diet included 20% corn starch which was replaced by the test ingredients. Fecal samples were collected on day (d) 20, 42, 62 and 86 to determine ATTF of NDF. On each collection day, 300 mL cecum content from 3-5 cannulated pigs was used as inoculum for a 72-hour *in vitro* fermentation of NDF fraction of ingredients in Ankom RF gas production system. ATTF data was analyzed with a general linear model in R. *In vitro* gas production data was analyzed with a non-linear curve fitting to generate coefficients i.e. intercept A, organic matter cumulative volume (OMCV) and Pearson correlation with ATTF data. Pigs fed soy hulls had highest ATTF (51.8%) while pigs fed rice hulls had the lowest (4.4%) ($P < 0.01$). Significant correlations were observed between *in vivo* ATTF and *in vitro* A ($P = 0.059$ or less) and OMCV ($P < 0.05$) per phase for all ingredients. The ATTF of NDF increased with age ($P < 0.01$), with the highest value at d86 (46.6%). In conclusion, data generated from *in vitro* NDF fermentation model highly correlated with *in vivo* ATTF in GF pigs. ATTF of NDF in the tested ingredients increased with age. **Key Words:** Growing-finishing pigs, NDF, Apparent total tract fermentability

**143 Investigating the impact of deoxynivalenol on digestive physiology and intestinal function in nursery pigs.** T. A. Crome*[1], D. J. Bloxham[2], and N. K. Gabler[1], [1]*Iowa State University, Ames, Iowa, United States*, [2]*Adisseo, USA, Alpharetta, GA, United States*. The mycotoxin deoxynivalenol (DON) can significantly reduce pig growth and health. This study evaluated the effects of DON on digestive physiology and intestinal function in nursery pigs using two experiments. Study 1 involved 200 newly weaned pigs (6.1 ± 1.02 kg) fed corn-soybean meal diets with either 0 (CON) or 3 ppm DON for 42 days (n=8 pens/treatment). Body weights and feed intake were recorded on days 0 and 42 to calculate ADG, ADFI, and Gain:Feed. From days 35–42, apparent total tract digestibility (ATTD) of dry matter (DM), nitrogen (N), and gross energy (GE) was determined. Pigs were necropsied to assess ileum morphology and markers of intestinal integrity and function. Study 2 involved 16 pigs (20.8 ± 4.59 kg) fed the same diets (n=8 pigs/treatment) for 35 days. On day 34, pigs were fasted overnight, re-fed ad libitum for four hours, and necropsied to evaluate gastric emptying and pH. In Study 1, DON reduced body weights (11%; $P = 0.001$), ADG (0.51 to 0.43 kg/d; $P < 0.001$), and ADFI (0.52 to 0.42 kg; $P = 0.003$) without affecting feed efficiency, ATTD coefficients, or ileum morphology. DON tended to increase intestinal permeability ($P = 0.08$), but transepithelial resistance was unaffected ($P = 0.497$). In Study 2, DON pigs consumed 22% less feed ($P = 0.016$) after fasting and retained less feed in their stomachs at four hours (103 vs. 71 g, $P = 0.015$), although meal retention (36 vs. 31%), gastric emptying rates (26 vs. 18 g/h), and stomach pH (2.87 vs. 2.56) were similar. In conclusion, DON reduced feed intake and growth in nursery pigs without affecting nutrient digestibility or gastric emptying rates. **Key Words:** mycotoxin, nursery, digestibility

**144 Effects of a multi-carbohydrase supplementation on digestive and metabolic utilization of energy in growing pigs.** Pierre Cozannet[1], Francis Amann Eugenio*[1], Maamer Jlali[1], Mark Giesemann[2], and Jean Noblet[3], [1]*Adisseo France SAS, ELISE - European Laboratory of Innovation Science & Expertise 20 rue Prosper Monnet, 69190, Saint Fons, France.*, [2]*Adisseo USA Inc, 4501 North Point Pkwy, Alpharetta, GA 30022, United States*, [3]*ex INRAe, INRAE, Rennes, France*. Net energy (NE) evaluation of feeds offers significant benefits for exogenous enzyme assessment in monogastrics nutrition. This study investigates the effect of a multi-carbohydrase (MC) on digestible (DE), metabolizable (ME) and NE in growing-finishing pigs fed a wheat-barley-peas-soybean meal-based diet. Six replicates of 2 littermate pigs with initial body weight of 48-50 kg were used. In each replicate, one pig received the control diet without MC (C diet) and the other one the control diet supplemented with MC (MC diet) The experiment was split into a 11 days adaptation period followed by a 10 days measurement period in a respiration chamber. Animals were provided 2 kg of feed per day, distributed in 3 meals. Feed intake, fecal output and heat production (HP) and its components were measured. Data were subject to variance analysis including block (n = 6) and enzyme (n = 2) as fixed effects. The apparent total tract digestibility (ATTD) of energy was not significantly improved (+0.7% unit) and the ME:DE ratio was slightly lower (-0.2% unit; P > 0.05) with MC supplementation. A detailed analysis of HP and its components showed that HP, adjusted for similar levels of physical activity and feed intake, tended to be lower in pigs fed the MC diet , leading to an increase of retained energy (+32 kJ/day/kg $BW^{0.60}$, $P <$ 0.05) and the efficiency of ME for NE (78.6 vs 77.6%, $P < 0.05$) and then a reduced thermic effect of feeding (-30 kJ/day/kg $BW^{0.60}$, $P <$ 0.05) in MC diet. Consequently, the ME and NE contents of MC diet were 0.12 and 0.23 MJ/kg DM ($P < 0.05$) higher. The present study suggests a better and higher evaluation of MC effect on energy value in a NE system compared with ME or DE systems in connection with a better account of the metabolic improvement associated with MC supplementation. Further work is required to identify the origin of energy saving associated with MC supplementation. **Key Words:** net energy, exogenous enzyme, metabolism

**145 Kinetics of protein digestion of weanling piglet measured in vitro for 27 feedstuffs.** D. Guillou*, C. Pineda Vadillo, and J. D'Amedor de Mollans, *Mixscience, Bruz, Bretagne, France*. After weaning, protein digestibility varies daily and differentially between individual piglets, making difficult the measurement of robust digestibility coefficients. An in vitro static model of the upper gastro-intestinal tract was constructed following the INFOGEST guidelines, to simulate protein digestion in oral, gastric and small intestine compartments of a 21 day-old weanling piglet. Digestion was performed in triplicate, with serial sampling of digesta, split into centrifugation pellet (PEL) and supernatant (SUP). The latter was ultra-filtered to obtain a permeate (PER) fraction of free amino acids and small peptides and a fraction of larger molecules retained on the filter (RET). Three time points were chosen to describe digestion: end of orogastric phase (0), after 30 min (30) and 300 min (300) of intestinal digestion. Twenty-seven protein-rich feedstuffs commonly used in swine feed in France were studied. Crude protein (CP) was analyzed in feedstuff, then at each sampling time in SUP and PER. CP in PEL and RET was calculated by difference based on weight. To describe digestion kinetics, CP was split in four parts: digested at time 0, additionally digested at time 30, additionally digested at time 300, and not digested. Hierarchical clustering was performed to study the data. Seven clusters were identified, based on the phase where more digestion took place. Among highly digestible protein sources, bovine plasma, egg powder and autolyzed yeast cell content belonged to the cluster of CP digested essentially in the orogastric phase. On the contrary, maize gluten showed a major contribution of the late digestion. High fiber oilseed meals (canola, sunflower), meals from expeller process and extruded full-fat soybean belonged to the cluster of incomplete CP digestion. Altogether, in vitro digestion kinetics of CP allows a novel approach for studying the equivalence of protein sources and level in the nutrition of weanling piglets. **Key Words:** weanling piglet, in vitro, digestion kinetics

**146 Standardized ileal amino acid digestibility of faba been, dehulled faba bean, peas, rapeseed meal, sunflower meal and three batches of soybean meal fed to growing pigs.** K. Blaabjerg*, S. K. Boldsen, P. Tybirk, N. M. Sloth, and U. P. Krogh, *Seges Innovation, Aarhus N, Denmark*. Standardized ileal digestibilities (SID) of amino acids (AA) in three batches of soybean meal and alternatives to soybean meal were determined, to enable inclusion of locally grown protein in pig diets without compromising accurate supply of digestible AA. Two experiments (Exp) were conducted. In each Exp, eleven pigs were ileal cannulated at a body weight (BW) of 33 kg. After a recovery period, the pigs were used in a Youden square design with five dietary groups and 8 periods. In Exp-I, pigs were fed a nitrogen-free diet (NF-1, n = 18) or one of four cornstarch-based diets with 32% standard soybean meal (SBM-1, n = 15), 64% faba bean (FB, n = 15), 43% rape seed meal (RSM, n = 16) or 78% peas (PEA, n = 15) as the sole AA source. In Exp-II, pigs were fed a nitrogen-free diet (NF-2, n = 20) or one of four cornstarch-based diets with 35% standard soybean meal (SBM-2, n = 16), 34% high protein soybean meal (SBM-3, n = 15), 60% dehulled faba bean (DFB, n = 13) or 46% sunflower meal (SFM, n = 15) as the sole AA source. Pigs were fed 43 g/kg BW and Yttrium was used as indigestible marker (0.4 g/kg). In each period, pigs were adapted to the diets for 5 days followed by two days of continuously ileal digesta collection for 9 hours per day. The SID of AA was calculated using average basal endogenous losses across experiments conducted with the same protocol and in the same facilities (average: 16.2 g AA/kg DM intake). Statistical analysis was conducted including dietary group and BW within dietary group as fixed effects and the pig and period as random effects. In Exp-I, SID of total AA was greatest ($P < 0.05$) for SBM-1 (82%), intermediate for PEA (77%) and FB (77%) and lowest for RSM (72%). In Exp-II, SID of total AA was greater in SBM-2 (90%) than in the remaining ingredients ($P < 0.05$), where SID of total AA was 86% for SBM-3, 83% for DFB, and 83% for SFM. In conclusion, SID of total AA of the three studied batches of SBM varied from 82% to 90%. SID of total AA of FB was 77% and 83% in DFB. **Key Words:** Ileal amino acid digestibility, protein sources, pigs.

**147 *In vitro* digested ingredients as substitute for ileal digesta in assessing protein fermentation potential in growing pigs.** H. Zhang*[1,2], J. Cone[1], A.K. Kies[3], W.H. Hendriks[1], and N. van der Wielen[4], [1]*Animal Nutrition Group, Department of Animal Sciences, Wageningen University & Research, Wageningen, The Netherlands*, [2]*State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China*, [3]*ArieKiesAdvies, Druten, The Netherlands*, [4]*Division of Human Nutrition and Health, Department of Agrotechnology and Food Sciences, Wageningen University & Research, Wageningen, The Netherlands*. Understanding hindgut protein fermentation is essential for enhancing gut health and nutrient utilization in pigs. Traditionally, ileal digesta from pigs fed diets with specific protein sources as the sole dietary protein is used to investigate fermentation *in vitro*. This study, however, examined the feasibility of using *in vitro* digested residues (IVDR) as a substitute for ileal digesta to assess the fermentation potential of protein sources. IVDR were prepared for several protein sources, including maize germ meal (MGM), cottonseed meal (CSM), rapeseed cake (RSC), rapeseed meal (RSM), peanut meal (PM), soybean meal (SBM), and sunflower meal (SFM). Samples containing 10 mg of nitrogen were incubated in sealed bottles with an excess of carbohydrates and faecal inoculum derived from 20 pigs fed commercial diets under standardized anaerobic conditions. Gas production (GP) was monitored over 48 h and analysed using a modified curve-fitting model. Significant differences in GP parameters, identified through Tukey's test, highlighted variations in nitrogen utilization by faecal microbiota across protein sources. SFM and SBM showed the highest maximum gas production rates ($R_{max}$) at 29.5 ± 0.6 mL/h and 28.0 ± 1.2 mL/h, respectively, while MGM exhibited the lowest rate at 17.3 ± 0.2 mL/h. Comparison of IVDR parameters with ileal digesta data from a previous study revealed a strong positive correlation between $R_{max}$ values ($R^2 = 0.85$, $P < 0.01$), indicating that IVDR can reliably predict the rate of protein hydrolysis in ileal digesta. However, GP potential ($GP_s$) demonstrated a weaker negative correlation ($R^2 = 0.39$, $P < 0.1$), possibly due to the narrow range of $GP_s$ values and the presence of endogenous proteins in ileal digesta. These findings suggest that IVDR can serve as a practical alternative to ileal digesta for evaluating the fermentation potential of protein sources, offering a useful

tool for studying fermentation dynamics and their implications for pig nutrition. **Key Words:** Protein fermentation, *in vitro* digestion, protein ingredients

**148 Increasing doses of a novel biosynthetic bacterial 6-phytase supplementation improves nutrient digestibility and growth performance in nursery pigs.** M Jlali* and S Ozbek, *Adisseo France S.A.S, Department of R&I in Monogastric Animal Nutrition, European Laboratory of Innovation, Science and Expertise, 69190 Saint-Fons, France*. This study aimed to investigate the efficacy of increasing doses of a novel biosynthetic bacterial 6-phytase on nutrient digestibility and growth performance in nursery piglets. Male weaned piglets (n = 192) with an initial body weight of 8.7 ± 1.1 kg were allotted to one of four treatments with 24 replicate pens (2 animals/pen) per treatment for 42 days. A basal corn-soybean meal-based diet (control diet) was formulated at 0.21% and 0.19% of standardized total tract digestibility (STTD) of P and 0.58% and 0.54% of calcium (Ca), in post-weaning 1 and 2 diets, respectively. Phytase was added to the control diet at 500, 1,000 or 1,500 phytase units (FTU)/kg diet. The diets were fed into 2 phases: post-weaning 1 and 2 from 1 to 14 and 15 to 42 days, respectively. Fecal samples were collected over 5 consecutive days from 12 pens per treatment during feeding post-weaning 2 phase for apparent total tract digestibility (ATTD) and phytate-P disappearance determination. Body weight (BW) and feed intake (FI) were measured at the end of each phase, and average daily gain (ADG), average daily feed intake (ADFI) and feed-to-gain (F:G) ratio were calculated for the whole experimental period. Increasing phytase supplementation linearly increased ($P < 0.001$) ATTD of dry matter (DM), nitrogen (N), crude fat, Ca, and energy. There was a tendency (P = 0.10) for quadratic effect of phytase supplementation on ATTD of Ca. Both linear and quadratic effects ($P < 0.001$) were observed for ATTD of P. ATTD of P was 45.47, 57.43, 71.18 and 71.86% with phytase at 0, 500, 1000 and 1,500 FTU/kg diet, respectively. Increasing phytase dose from 0 to 1,500 FTU/kg diet linearly increased (P = 0.006) the overall ADFI (+7.2%) and ADG (+10.7%) and decreased ($P < 0.001$) F:G ratio (-3.2%). These findings confirmed that supplementation of nursery pigs' diets with the novel biosynthetic bacterial 6-phytase significantly improved the nutrient digestibility and growth performance in a dose-response manner. **Key Words:** Efficacy, phytase, nursery pigs

**149 Difference in jejunal transcriptomic profile between low and high feed conversion ratio grower-finisher gilts fed a diet with a standard crude protein level.** M.E. van der Heide*[1], A.R. Williams[2], J.V. Nørgaard[1], and J.G. Madsen[2], [1]*Department of Animal and Veterinary Sciences, Aarhus University, Tjele, Denmark*, [2]*Department of Veterinary and Animal Sciences, University of Copenhagen, Frederiksberg, Denmark*. Understanding the biological mechanisms behind feed conversion ratio (FCR, feed:gain) in growing-finishing pigs is of great importance to lower nutrient excretion and carbon footprint. Sixty gilts were divided between three dietary treatments (low; 14.8%, standard; 15.2%, high; 15.8% crude protein (CP)) during a one phase feeding regime from 30 kg to 95 kg BW. Individual feed intake was measured and thus all growth performance parameters were estimated using pig as observational unit. Four weeks prior to slaughter 12 pigs per dietary treatment were categorized as having either low (n = 6) or high (n = 6) FCR. Pigs were euthanized and 150 mg jejunal tissue was sampled and snap frozen in liquid nitrogen. Ribonucleic acid was extracted and subjected to a transcriptomic analysis. Within the standard CP level dietary treatment 58 genes were differentially expressed (Adj. $P < 0.05$) with two being upregulated in high FCR pigs compared to low FCR pigs, and 56 being more highly expressed in low FCR pigs. Notably, gene pathways related to selenoaminoacid metabolism, co-translational target to membrane, rRNA processing, translation elongation and translation initiation were enriched ($P < 0.05$) in high compared with low FCR pigs, while pathways related to elastic fiber formation, voltage gated potassium channels, smooth muscle contraction, cell junction organization, adherens junction organizations were enriched ($P < 0.05$) in low compared with high FCR pigs. In contrast, only minor gene changes were evident in pigs on the high CP diet, and no genes were regulated in the low CP diet. In conclusion, the effect of FCR on the transcriptomic response was dependent on dietary crude protein level, where the enriched biological pathways in high

compared with low individuals were generally associated with protein metabolism and cell proliferation, while pathways involved in extracellular matrix organization and cell-cell communication were generally enriched in low compared with high individuals. **Key Words:** Transcriptomics, Feed conversion ratio, Jejunum

**150 Novel ingestible sensor methodology for continuous measurements in the gastrointestinal tract.** R. Minderhoud*[1,3], A. Even[4,5], T. Torfs[6], F. Leonardi[4,5], A. van Heusden[4,5], R. Sijabat[4,5], D. Firfilionis[4,5], E. Capuano[2], G. Hooiveld[1], and S. de Vries[3], [1]*Division of Human Nutrition and Health, Wageningen University & Research, Wageningen, The Netherlands*, [2]*Food Quality and Design Group, Wageningen University & Research, Wageningen, The Netherlands*, [3]*Animal Nutrition Group, Wageningen University & Research, Wageningen, The Netherlands*, [4]*OnePlanet Research Center, Wageningen, The Netherlands*, [5]*imec, Wageningen, The Netherlands*, [6]*imec, Leuven, Belgium*. Studying conditions in the gastrointestinal tract (GIT) is challenging due to its length and inaccessibility. Current methods such as the slaughter technique and T-cannulas (in animals) and endoscopies (in humans) are invasive, while alternative biospecimen like fecal samples only reflect the last part of the GIT. To overcome these challenges, a highly miniaturized ingestible was developed that can measure pH, temperature, and oxidation-reduction potential (ORP) throughout the GIT at 20 second intervals. Redox balance is important for maintaining the intestinal barrier and facilitating interactions between host, immune system, and GIT microbiota. To explore the usability, safety, and ability of ingestible sensors to monitor GI conditions and transit non-invasively, they were tested in 7 pigs. Every pig was given one ingestible. Two pigs were given a second ingestible. In total, data from 9 ingestibles were obtained. The sensor data were divided into stomach, small intestine, and large intestine based on pH profiles following common used thresholds in literature. The data revealed that ORP decreased from an oxidative environment with values ranging between +250 mV and -50 mV in the stomach, to -50 mV and -250 mV in the small intestine, and a strongly reducing environment in the large intestine below -300 mV. For four out of nine ingestibles large intestine entry could not be defined solely based on pH profiles, while ORP did show a large change from small intestine to large intestine (100 mV to -325 mV). Transit time of the ingestibles varied from 2 to ≥ 7 days. In conclusion, this ingestible sensor was able to measure temperature, pH, and ORP continuously inside the GIT for multiple days. The ORP sensor provided novel data of *in vivo* redox potential along the GIT and also improved localizing the ingestible, compared to pH sensor only. This innovative method can enhance our understanding of nutritional interventions and GIT-linked diseases in both pigs and humans. **Key Words:** ingestible sensor, redox potential, pH

**151 Comparison of the dual isotope tracer approach with oro-ileal balance method for determination of amino acid digestibility in cannulated pigs.** N van der Wielen*[1,2], S de Vries[1], N Khodorova[3], J Calvez[3], I Minussi[1], W Gerrits[1], C Gaudichon[3], D Tome[3], and M Mensink[2], [1]*Animal Nutrition group. Wageningen University & Research. The Netherlands.*, [2]*Division of Human Nutrition. Wageningen University and Research. The Netherlands.*, [3]*UMR PNCA, AgroParisTech, INRA, Université Paris-Saclay, 75005 Paris, France.* The dual isotope tracer approach was developed as a less invasive alternative for the measurement of standardized ileal digestibility (SID) of amino acid (AA) in humans. *Objective:* This study aimed to validate the dual isotope tracer approach with the oro-ileal balance method in growing pigs. Eight gilts (29.7±1.6 kg) were fitted with jugular catheters and ileal T-cannulas. At the test day, feed, containing intrinsically labeled $^{15}$N-milk protein (147g/kg DM) and $^{13}$C-spirulina (0.019g/kg DM), was provided every half hour, during 240 minutes. Ileal digesta and ten jugular blood samples were collected between 0-540 minutes. Digesta samples, pooled from 270 to 540 minutes after ingestion of the first meal, were analyzed for isotopic enrichment, amino acid, and titanium concentrations for SID calculations according to oro-ileal balance method. Serum AA isotopic enrichment was measured at baseline and averaged between 240, 300 and 360 minutes after first ingestion for dual stable isotope tracer calculations. Methods were compared using paired t-test and Spearman correlation. Using the oro-ileal balance method the average SID of AA from milk protein

concentrate was 97.8±0.59% and of spirulina 81.5 ± 2.44% (mean ± SD). Lysine digestibility was specifically evaluated, as it does not transaminate. Lysine digestibility of milk protein concentrate calculated according to the dual isotope tracer approach was 88.9 ± 8.35%, 9 % point lower than the value obtained with the oro-ileal method (98.1 ± 0.36, $P$ = 0.04). Moreover, digestibility values of lysine across animals did not correlate between methods ($r$ = -0.09, $P$ = 0.87). This study observed differences between the dual isotope tracer approach and oro-ileal balance method for estimating lysine digestibility under the current experimental conditions. Considering the use of $^{15}$N protein, conclusions on other AA that do not transaminate could not be drawn. **Key Words:** Dual isotope tracer approach, amino acid digestibility, SID

**152 Standardized ileal amino acid digestibility increased with body weight of growing pigs fed faba beans, dehulled faba beans and peas, while unaffected by body weight when fed soybean meal, sunflower meal and rapeseed meal.** U. P. Krogh*, S. K. Boldsen, P. Tybirk, N. M. Sloth, and K. Blaabjerg, *Seges Innovation, Aarhus N, Denmark*. The objective was to investigate how bodyweight (BW) of pigs between 40 and 60 kg affect their ability to digest amino acids (AA) from different protein sources. Two experiments (Exp) were conducted. In each Exp, eleven growing pigs were ileal cannulated at a BW of 33 kg. After a recovery period, the pigs were used in a Youden square design with five groups and 8 periods. In Exp-I, pigs were fed a nitrogen-free diet (NF-1, n = 18) or one of four cornstarch-based diets with either standard soybean meal (SBM-1, n = 15), faba bean (FB, n = 15), rape seed meal (RSM, n = 16) or peas (PEA, n = 15) as the sole amino acid (AA) source. In Exp-II, pigs were fed a nitrogen-free diet (NF-2, n = 20) or one of four cornstarch-based diets with either standard soybean meal (SBM-2, n = 16), high protein soybean meal (SBM-3, n = 15), dehulled faba bean (DFB, n = 15) or sunflower meal (SFM, n = 15) as the sole AA source. Pigs were fed 43 g/kg BW and Yttrium was used as indigestible marker (0.4 g/kg). In each period, pigs were adapted to the diets for 5 days followed by two days of continuously ileal digesta collection for 9 hours per day. The standardized ileal digestibility (SID) of AA was calculated using average basal endogenous losses (16.2 g AA/kg DM intake) from the two Exp and a previous study conducted in the same facilities (n = 7). Statistical analysis was conducted in R and included group and BW within group as fixed effects and pigs and period as random effects. The initial and final BW of pigs was 36 and 57 kg in Exp-I and 38 and 64 kg in Exp-II. The SID AA increased with BW by 0.27 %/kg in DFB, 0.35 %/kg in FB and 0.55 %/kg DFB ($P$ < 0.05). In contrast, SID of total AA was not affected by BW when fed SBM-1, SBM-2, SBM-3, RSM and SFM ($P$ > 0.10), likely because these sources were exposed to a higher degree of processing. In conclusion, BW did not affect SID AA of processed oil by-products, while SID of AA increased linearly with BW in growing pigs fed PEA, FB and DFB. **Key Words:** Ileal amino acids digestibility, protein sources, pigs.

**153 Fat digestibility is reduced and may be overestimated in by-product based diets used for circular food production.** P. Bikker*[1], R. Gerritsen[2], M. van Helvoort[3], P. Pluk[4], M. Schop[5], and E. Royer[1], [1]*Wageningen University & Research, Wageningen Livestock Research, Wageningen, the Netherlands*, [2]*ForFarmers, Lochem, the Netherlands*, [3]*De Heus Animal Nutrition, Ede, the Netherlands*, [4]*Cargill Animal Nutrition, Velddriel, the Netherlands*, [5]*Agrifirm, the Netherlands*. Future pig diets in Europe may contain less cereal grains and soybean meal (SBM) and more regional protein sources and by-products as alternative ingredients to improve circularity of food production. Fat and non-starch polysaccharides (NSP) are major energy sources in these diets. Because of their potential interaction, results of 5 recent studies were used to evaluate apparent total tract digestibility (ATTD) of fat and NSP and to compare values with those based on the Dutch CVB feed table. In 3 experiments in weaned pigs' complete replacement of SBM by other oil seed meals or legume seeds, and replacement of wheat by bakery products or by-products of maize and wheat processing was evaluated. In 2 experiments in growing finishing (GF) pigs replacement of both wheat and SBM by bakery products and oil seed meal, legume seeds or by-products of maize and wheat processing was studied. Dietary NSP content varied between 140 and 260 g/kg. Each study comprised 4 to 8 treatments with 8-16 replicates (pens) per treatment. Faeces were collected by grab sampling to determine ATTD. A post-hoc

evaluation was conducted per experiment using ANOVA with diet as fixed factor. In all studies, ATTD of fat and NSP was affected ($P < 0.05$) by the inclusion of alternative feed ingredients, as was expected based on table values. With the exception of the bakery product diet in GF pigs, the ATTD of crude fat was 4-10 %-units below expected based on table values for the diets with legume seeds and by-products of cereal grains and oil seeds. This effect was larger in weaned pigs than in GF pigs. Results suggest that alternative ingredients may reduce fat digestion, presumably due to the non-starch polysaccharide fraction affecting passage rate, viscosity, enzyme accessibility, and bile acid binding in the digestive tract. In conclusion, diets with a high proportion of alternative ingredients may have reduced ATTD of fat and lead to overestimation of fat digestibility based on table values. **Key Words:** Digestibility, Fat, By-products

**154 The chalk limestone provides more digestible Ca and enables phytase to release more P than marble limestone in pigs.** H X Zhai[1], S K Wang*[1], J C Zhang[1], Z Z Wang[1], and J B Liu[2], [1]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Bazhou, China*, [2]*School of Life Science and Engineering, Southwest University of Science and Technology, Mianyang, China*. The objective of this study was to assess the impact of limestone quality (marble: low solubility vs. chalk: high solubility) on the digestible Ca and P equivalence of phytase in nursery pigs. This study included 8 experimental diets arranged in a 2 × 4 factorial design with 2 limestone sources and 4 concentrations of a novel phytase (0, 1,000, 2,000 and 4,000 FYT/kg of feed). A total of 24 barrows (initial body weight 10.3 ± 0.4 kg) were housed individually in metabolism cages and each diet was fed to 8 barrows. All diets contained 3 g/kg TiO2, and fecal samples were collected for 5 days following a 5-day adaptation period. Limestone solubility was assessed in a pH 3 solution buffered with 3 M glycine. The data were analyzed using ANOVA. The results indicated no significant interaction between phytase dose and limestone quality for apparent total tract digestibility (ATTD) of Ca. The ATTD of Ca increased exponentially with increasing phytase doses ($P < 0.01$). The diets with chalk limestone resulted in significantly higher ATTD of Ca than the diets with marble limestone, aligning with the higher in vitro solubility of chalk limestone. A significant interaction between phytase dose and limestone quality was observed for the ATTD of P. In the absence of phytase, diets with chalk limestone showed lower ATTD of P compared to those with marble limestone. However, phytase supplementation reversed this trend. Within each source of limestone, the ATTD of P increased exponentially with increasing phytase doses ($P < 0.01$). There was no significant effect of phytase dose and limestone quality on digestible Ca equivalence. However, the digestible P equivalence increased linearly with increasing phytase doses ($P < 0.01$) and was significantly higher in pigs fed chalk limestone compared to those fed marble limestone. In conclusion, the chalk limestone should be recommended for pigs not only for its higher Ca digestibility but also for its greater response to phytase supplementation. **Key Words:** Limestone, Phosphorus, Phytase

**155 Comparison of recommendations for standardized total tract digestible Ca and total Ca requirments in growing-finishing pigs fed diets with or without phytase.** H X Zhai*[1], S K Wang[1], J C Zhang[1], Z Z Wang[1], and J B Liu[2], [1]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Bazhou, China*, [2]*School of Life Science and Engineering, Southwest University of Science and Technology, Mianyang, China*. Different calcium (Ca) requirements for growing-finishing pigs have been recommended. The objective of this study was to compare the standardized total tract digestible (STTD) Ca recommendations by Lee et al. (2023) and total Ca recommendations by NRC (2012) for growing-finishing pigs fed diets with or without a novel phytase. Six experimental diets were arranged in a 3 × 2 factorial design, with three Ca recommendations (STTD Ca recommendations for optimal growth performance and bone mineralization, and total Ca recommendations) and two phytase levels [0 and 1,000 FYT/kg of feed (equivalent to 0.15% digestible phosphorus (P) and 0.11% digestible Ca)]. Each diet was fed to eight pens of six pigs each, totaling 288 pigs (initial body weight 25.8 ± 2.8 kg) over an 84-day period across three growth stages. At the end of the study, one pig from each pen was sacrificed for the collection of the right tibia. The data were analyzed using ANOVA. The results showed no significant effect of Ca

recommendations, phytase, or their interaction on the growth performance of pigs. Additionally, there was no significant interaction between Ca recommendations and phytase regarding bone mineralization. The digestible Ca recommendation for optimal bone mineralization resulted in heavier weights of dry bone and bone ash, and a higher percentage of bone ash than the digestible Ca recommendation for optimal growth performance ($P < 0.01$). These parameters were intermediate for the total Ca recommendation treatment. There was no significant effect of Ca recommendations on the percentage of bone Ca and P. Furthermore, no significant difference in any of the bone parameters was observed between the diets with and without phytase. In conclusion, this study demonstrates that there is no difference in growth performance between different Ca recommendations. However, optimal bone mineralization can be achieved by using the digestible Ca recommendation specifically designed for this purpose. **Key Words:** Calcium requirement, Phosphorus, Phytase

**156 Effects of Bacillus-based probiotic application to sows on sow and suckling pig performance under heat stress.** K.P. Kinsley*[1] and L. Hübertz Birch Hansen[2], [1]*Novonesis, West Allis, WI, United States*, [2]*Novonesis, Lyngby, Denmark*. Heat stress in sows disrupts lactation, increases oxidative stress, and reduces performance. We hypothesize that *Bacillus*-based probiotics (BBP) may promote amino acid breakdown into antioxidant precursors, reducing free radical damage. This study aimed to assess whether BBP supplementation to sows can mitigate heat stress and improve lactation performance. Thirty-two, parity 2 and 3 bred sows were acquired in May with late July anticipated farrowing dates. Sows were randomly allotted to one of two treatments (+/- BBP consisting of *B. licheniformis* 809A and *B. subtilis* 810A). A daily top dress application a BBP or control (CON) product was given from start gestation to weaning (21d post-farrow). No attempts were made to abate heat in the gestation facility outside of minimum ventilation fans. In the farrowing unit, temperature set points were set to keep minimum temperatures at or above 27°C per 24-hour period. Sows and litters were clinically assessed daily for indicators of heat stress – respiratory distress (0 = normal to 2 = severe respiratory distress, apparent dyspnea), depressed attitudes (0 = normal to 2 clinical depression, listless, will not rise), and daily inappetence level (0 = normal to 3 = complete feed refusal). Sow and piglet serum samples were collected at farrowing and weaning to analyze antioxidant markers. During the farrowing period, there was no difference in sows exhibiting signs of heat stress (measured by days where more than one clinical metric score was > 0, $P > 0.05$). However, inappetence was a leading indicator with 15.1% of all CON and 8.1% of BBP lactation days experiencing complete feed refusal ($P < 0.01$). Additionally, individual pig wean weight (5.0 vs. 5.6 kg, $P < 0.001$) and birth to wean gain (3.4 vs. 3.8 kg, $P < 0.001$) were greater in the BBP treatment despite no difference in pig birth weights. In conclusion, BBP supported lactation performance under heat stress conditions. **Key Words:** Bacillus, Litter, Stress

**157 Variable dietary calcium to phosphorous ratios and microbial phytase did not alter portal vein profiles of blood acid-base balance, blood gases, and electrolyte concentrations in pigs during a 10-hour post-absorption phase.** A.P.U. García[1,2], T.D. Crenshaw[3], A. Narcy[4], P. Schlegel[5], M-P. Létourneau-Montminy[2], and D.B. Dalto*[1], [1]*Agriculture and Agri-Food Canada, Sherbrooke R&D Centre, Sherbrooke, Quebec, Canada*, [2]*Université Laval, Department of Animal Science, Quebec, Quebec, Canada*, [3]*University of Wisconsin, Department of Animal and Dairy Sciences, Madison, Wisconsin, United States*, [4]*INRAE, Université de Tours, Nouzilly, Centre-Val de Loire, France*, [5]*Agroscope, Swine Research Unit, Posieux, Hauterive, Switzerland*. This study compared different dietary calcium (Ca) to standardized total tract digestible phosphorous (STTD P) ratios and the use of microbial phytase on blood gases, electrolytes, and acid-base balance in pigs. Seven growing pigs (42.9 ± 2.2 kg) were equipped with portal vein catheters and assigned to five meals with different Ca:STTD P ratios: CTR - 2.6 Ca:STTD P ratio; C-Phy - 2.6 Ca:STTD P ratio + 750 phytase units (FTU)/kg; C-Phy+ - 2.6 Ca:STTD P ratio + 2,500 FTU/kg; LCa - 1.5 Ca:STTD P ratio; and L-Phy - 1.5 Ca:STTD P ratio + 750 FTU/kg. Portal blood samples were collected (2 experimental days per week) at 0, 2, 6, and 10 hours (hr) post-meal and analyzed for pH, partial

pressure of carbon dioxide (pCO2), partial pressure of oxygen (pO2), bicarbonate (HCO3), base excess (BE), sodium (Na), potassium (K), ionized calcium (iCa), and glucose (GLU) concentrations. Data were analyzed as a 5x5 Latin Square design with repeated measures over time. Differences were considered significant at $P \leq 0.05$ and tendencies at $0.05 < P \leq 0.10$. Most parameters were not impacted by treatments ($P \geq 0.16$) except that C-Phy tended ($P > 0.07$) to increase GLU compared to C-Phy+. Although no treatment x time interaction was detected ($P \geq 0.16$), the multiple comparison analysis shows that 10 hr after meal iCa was highest for C-Phy and lowest for LCa and L-Phy. Time effect was detected for most parameters ($P \leq 0.02$). Blood pH and K were highest 2 hr after meal and lowest at 10 hr. Blood BE, HCO3 and GLU were lowest at 0 and highest at 2 hr after meal. Blood Na was highest at 0 and lowest from 2 hr after meal whereas pCO2 was lowest at 0 and highest from 2 hr after meal. Blood iCa was lowest 2 hr after meal but by 10 hr returned to baseline (0 hr). In conclusion, there are remarkable changes in blood gases, electrolytes, and acid-base balance during 10 hr after meal. Different Ca:STTD P ratios did not impact the blood acid-base balance despite the lower iCa in low Ca:P ratio diets. **Key Words:** calcium:phosphorous ratio phytase portal vein

**158 Comparison of P digestibility among magnesium phosphate, monocalcium phosphate, and monosodium phosphate at different Mg levels fed to piglets.** N. Aubertin*[1], B. Ribeiro[1], M. Poujol[1], and V. Lagos[2], [1]*Phosphea, Dinard, France*, [2]*Schothorst Feed Research, Lelystad, The Netherlands*. This piglet study aimed to compare the apparent total tract digestibility (ATTD) of P of monocalcium phosphate (MCP), monosodium phosphate (MSP), and magnesium phosphate (MgP), and to evaluate the effect of dietary Mg level on P digestibility. Thirty weaned male piglets (8.50±2.0 kg, 30 days old) were assigned to a randomized complete block design with 5 experimental diets and 6 replicates per diet. Pigs were housed individually in metabolic cages and fed semi-ad libitum from day 17-25. The experimental diets were offered after a 7-day period with a common weaner diet. A semi-synthetic basal mix low in P and Mg was formulated and split into 5 batches to which the P source, limestone, and diamol were added. All diets had the same net energy, digestible Lys, Ca, and digestible P (3.06 g/kg), and included 0.5% $TiO_2$. Diets were formulated as a 2×2+1 factorial design with two P sources (MCP or MgP), two Mg levels (2.0 or 5.3 g/kg, provided by MgO or MgP), and a MSP, low Mg diet. The high Mg diets contained only MgP or MCP+MgO. The low Mg diets contained MSP+MgO, MCP+MgO, or MgP+MCP. Feces were collected from day 20-25. The ATTD of P in P sources was calculated by the difference procedure and the standardized total tract digestibility (STTD) of P was estimated based on dry matter intake. The three P sources were compared at low Mg level, and the main effects and interaction between P source and Mg level were evaluated. At low Mg level, MSP (94.1%) had the highest ($P < 0.05$) STTD of P, with no differences between MgP (81.9%) and MCP (83.4%) (SEM=2.61%). There was no interaction between P source and Mg level, but high Mg levels reduced (-10.4%; $P < 0.05$) the STTD of P in both P sources. The STTD of P in MgP and MCP at high Mg level was 73.8 and 70.6%, respectively, but no main effect of P source was observed. In conclusion, the use of high Mg levels in piglet feed decreases P digestibility regardless of P source, but there is no difference in P digestibility between MgP and MCP. **Key Words:** Digestibility, magnesium phosphate, piglets

**159 Phytase appeared to counterbalance the inimical effect on N balance of acidosis associated with displacing calcium carbonate with calcium chloride in grower pigs.** H Zhai*[1], E Perez-Calvo[2], S K Wang[1], J C Zhang[1], Z Z Wang[1], and J B Liu[3], [1]*dsm-firmenich, Animal Nutrition and Health, R&D Center, Bazhou, China*, [2]*dsm-firmenich, Animal Nutrition and Health, Kaiseraugst, Switzerland*, [3]*School of Life Science and Engineering, Southwest University of Science and Technology, Mianyang, China*. The objective of this study was to evaluate the effects of Ca source and phytase on the balance of Ca, Cl, and P and the chemistry of plasma and urine. There were 6 experimental diets arranged as a 3 × 2 factorial with 3 Ca sources [0.8% limestone (35.7% Ca), 0.8% $CaCl_2$, or their 1:1 mixture] and 2 phytase levels (0, and 1,000 FYT/kg). Each diet was fed to 8 barrows individually housed in metabolism cages. Fecal and urinary samples were quantitatively collected

for 5 days after an adaptation period of 5 days. The data was analyzed using ANOVA. The results showed that the plasma concentration of glucose was higher in pigs fed $CaCl_2$ than in pigs fed only limestone ($P < 0.05$). The supplemental phytase increased the plasma concentration of P but reduced the plasma concentrations of Ca and blood urea nitrogen ($P < 0.05$). Urinary pH was lower in pigs fed only $CaCl_2$ than other pigs ($P < 0.05$). The supplementation of phytase reduced urinary concentration of Ca ($P < 0.05$), and tended to reduce urinary concentration of N and to increase urinary concentration of P ($P < 0.10$). Increasing inclusion of $CaCl_2$ in place of limestone resulted in increases in apparent total tract digestibility and retention of Ca and P. The Cl intake, output through feces and urine, and retention, increased with increasing levels of $CaCl_2$ ($P < 0.05$). Phytase decreased Ca and P output through feces and increased Ca and P retention ($P < 0.05$). There was a decrease in retention of CP in diet including only $CaCl_2$ compared to other diets in the absence of phytase, which contrasts with an increase in the presence of phytase. In conclusion, the current study shows that $CaCl_2$ has a higher bioavailability of Ca than limestone. However, $CaCl_2$ can disrupt the dietary electrolyte balance with the consequence of reduced urinary pH. In addition, phytase appeared to be able to ameliorate the inimical effect of acidosis associated with $CaCl_2$ supplementation on N balance. **Key Words:** Acidosis, Calcium chloride, Phytase

**160 Metabolic interactions in weaned piglets: effects of dietary zinc source and level.** Jonathan Riedmüller[1], Wilfried Vahjen[1], Jamil Faccin[2], Alessandra Rigo Monteiro*[3], Joel DeRouchey[2], Jordan Gebhardt[2], Robert Goodband[2], Jason Woodwort[2], Mike Tokach[2], and Jürgen Zentek[2], [1]*Freie Universität Berlin, Berlin, Germany*, [2]*Kansas State University, Manhattan, USA*, [3]*Animine Precision Minerals*, *Annecy, France*. This study examined the metabolic interactions between zinc oxide sources and other minerals in weaned piglets. A total of 240 weanling barrows (DNA 200 × 400, initially 5.9 ± 0.05 kg) were randomly assigned to 4 treatments (12 pens/treatment; 5 pigs/pen). The experiment consisted of phase 1 (d 0 to 10), phase 2 (d 11 to 24), and phase 3 (d 25 to 38). Treatments consisted of a negative control (NC; 150 ppm of Zn from HZ; HiZox®, a ZnO source with small aggregates and agglomerate particles); positive control (PC) containing 3,000 ppm (phase 1) and 2,000 ppm (phase 2) of Zn provided by standard ZnO (Maximo®720; a ZnO source with large and dense particles); LowHZ containing 500 ppm (phase 1) and 300 ppm (phase 2) of Zn from HZ; HiHZ containing 800 ppm (phase 1) and 500 ppm (phase 2) of Zn from HZ. In phase 3, all pigs were fed NC diet. At d 10, 23 and 38, blood was collected. At d 23, one pig per pen was euthanized and liver samples were taken. Data were analyzed using the GLIMMIX procedure of SAS. A p-value ≤ 0.05 was considered to indicate significant differences. For the experimental period (d 0 to 24), ADG ($P = 0.007$) and ADFI ($P = 0.02$) increased (quadratic) as HZ increased. Analysis of mineral content in liver and serum showed no treatment effect on Ca and P. Iron concentration in the liver (average of 129 mg/kg) and serum (average 0.135 mg/l) at d 23 were not affected by treatments either. As expected, serum and hepatic Zn showed higher values with the increase in dietary Zn ($P < 0.01$). After reducing zinc concentration to 150 ppm for all animals in phase 3, NC and HZ treatments (average serum Zn = 0.104 mg/l) showed no differences, but remained higher for the PC (0.14 mg/l; $P < 0.01$). A lower Mn concentration in liver ($P < 0.01$) was observed when pigs were fed high Zn levels. In conclusion, zinc supplementation did not affect Ca, P and Fe status, but seems to decrease Mn status, while increasing hepatic and serum Zn in post weaning piglets. **Key Words:** Minerals, Swine, ZnO

**161 Extruded Corn and β-Mannanase addition reveal comparable growth rate, nutrient utilization, and reduced fecal score in growing pigs.** K.J. Lee*[1,2], V. Sampath[1,2], and I.H. Kim[1,2], [1]*Department of Animal Biotechnology, Dankook University, Cheonan, SouthKorea*, [2]*Smart Animal Bio Institute, Cheonan, SouthKorea*. The focal aim of this experiment was to investigate the comparative effect of β-mannanase and extruded corn on growing pigs' performance. A total of 84 [Duroc × (Landrace × Yorkshire)] pigs with a mean body weight of 29.20 kg were used in the 42-days trial. Pigs were randomly allotted one of three treatments, and the test treatments were: CON (corn soybean basal diet, non-extruded), TRT1 (CON plus 0.1 % β-

mannanase), and TRT2 (6% extruded corn). There were 7 replicates/trt with 4 pigs (2 gilts and 2 barrows)/pen. Growth performance [body weight (BW), average daily gain (ADG), average daily feed intake (ADFI), and gain to feed ratio (G: F)] were measured at days 1, 5, and 42. Nutrient digestibility [dry matter (DM), nitrogen (N), and energy (E)] and fecal score were measured at the end of d42. All data were analyzed using the GLM procedure of SAS. Duncan analyses were performed, and the significant difference was decided by comparing the means. $P < 0.05$ was considered statistically significant. Pigs fed diets containing 0.1% β-mannanase with non-extruded corn and 6% extruded corn showed a significant increase ($P < 0.05$) in body weight (BW) at the end of day 42 compared to those fed CON diet. Also, during day 5, 42, and the overall experimental period the daily gain and gain to feed ratio of both treatments' group pigs were ($P < 0.05$) increased significantly. Additionally, pigs fed extruded corn exhibited significantly higher ($P < 0.05$) DM and N digestibility, along with a lower diarrhea score, compared to the CON and TRT 1 group. In summary, we infer that feeding a non-extruded corn diet supplemented with β-mannanase, as well as an extruded corn diet would be more effective in improving growing pigs performance than feeding a non-extruded corn diet alone. **Key Words:** β-mannanase, extruded corn, nutrient digestibility

**162 Enhanced bioavailability and stress resilience of a combined phosphorus and magnesium source in an *in vitro* pig model.** A. Juanchich[1], E. Dupuis[1], B. Ribeiro[2], N. Aubertin[2], T. Chalvon-Demersay[1], and E. Coudert*[1], [1]*Centre Mondial de l'Innovation Roullier, Saint-Malo, Bretagne, FRANCE*, [2]*PHOSPHEA, Dinard, Bretagne, FRANCE*. Minerals' availability is critical for pig nutrition, supporting optimal health, growth, and productivity by enhancing absorption and minimizing excretion. This study evaluated a source of magnesium phosphate (MgP) in comparison to monocalcium phosphate (MCP) combined with magnesium oxide (MgO). The investigation focused on minerals bioavailability, response of cells to stress and molecular mechanisms activation through *in vitro* experiments. Phosphorus and magnesium solubilization were assessed using an *in vitro* digestion model. Absorption rates were measured with a Caco2 uptake assay. Viability assays were performed under both normal and stress -induced by increasing temperature up to 45°C- conditions. RNA levels of key genes associated with Mg transport, membrane integrity, and stress response was measured with quantitative PCR. Results were subjected to statistical analyses (ANOVA, Kruskal-Wallis test) using the R4.3.0 software. Magnesium phosphate significantly increased Mg solubilization and absorption rates compared to MCP+MgO ($P < 0.01$). Under normal conditions, MgP-treated cells demonstrated a higher viability compared to control cells and MCP-treated cells, indicating a protective effect ($P < 0.05$). Under heat stress, MgP-treated cells maintained higher viability compared to MCP-treated cells ($P < 0.05$) and superior antioxidant capacity ($P < 0.01$). At the molecular level, MgP enhanced magnesium transport by upregulating TRMP6 ($P < 0.05$) and SLC41A3 ($P < 0.10$) expression and tended to improve membrane integrity through increased CLDN1 expression ($P < 0.10$). Additionally, MgP regulated oxidative stress pathways by maintaining GPX2 expression at control levels, compared to MCP ($P < 0.05$). In conclusion, MgP demonstrates superior Mg bioavailability, enhanced cellular resilience, and enhanced activation of pathways for Mg transport and membrane stability. These findings need to be confirmed with *in vivo* trials to position MgP as a promising alternative for pig nutrition. **Key Words:** Phosphorus, Magnesium, Bioavailability

**163 Empowering Hyper-Prolific Sows: Can Enhanced Pen Design with Simulated Udders Improve Piglet Growth in their Early Life?** Christina Larsen*[1], Vivi Aaresturp Moustsen[2], Kimmie Kyed Lyderik[1], and Johannes Guldmann Madsen[1], [1]*University of Copenhagen, Copenhagen, Denmark*, [2]*SEGES Innovation, Aarhus N, Denmark*. Hyper-prolific sows give birth to large litters resulting in a greater variation in birth weights and increased reliance on nursing sows. This study aimed to optimize expanded farrowing pen space, as recommended by the European Food Safety Authority (EFSA), by supporting loose-housed sows in rearing litter sizes exceeding the functional teat capacity. The objective was to enhance piglet growth while maintaining production efficiency. At farrowing, 66 litters were randomly assigned to either a control group (CON; n = 33) or a treatment group (TRT; n = 33), including only piglets weighing above 750 g at birth. From farrowing to

weaning, CON sows reared 16 piglets per litter, while TRT sows reared 20. From day one to four post-partum, a confinement setup was installed in the creep area of pens housing TRT sows and litters. Repeatedly during this period, the four smallest piglets in each litter, based on birth weight, were confined and given access to milk replacer for the first 6 hours. This was followed by the next four smallest piglets for another 6 hours, and the four largest piglets were confined overnight for 12 hours. Piglets in the CON group were reared by the sow without intervention. Individual piglet body weight was recorded daily from day one to four, with additional growth recordings on days 11, 18, and 25. When correcting for litter size, total litter weight did not differ day one to four post-partum ($P > 0.05$). However, the CON group exhibited significantly greater litter weight on day 11, 18, and 25 ($P < 0.01$). Average daily gain was significantly higher in the CON group ($P = 0.04$; 89.3 vs. 75.6 g/d) day one to four. The confinement setup might assist hyper-prolific sows in rearing large litters, however, no direct benefits on growth performance were observed. This indicates a need for strategies supporting these piglets during the entire suckling period. **Key Words:** Artifical udder, Growth, Large litters

**164 Tracing emptying of fibres differing in physicochemical properties using the Human Gastric Simulator: comparison with *in vivo* gastric retention times in pigs.** Corentin Lannuzel[1], Sonja de Vries*[1], Walter J.J. Gerrits[1], and Gail M. Bornhorst[2,3], [1]*Wageningen University & Research, Animal Nutrition, Wageningen, the Netherlands*, [2]*Department of Biological and Agricultural Engineering, University of California, Davis, CA, USA*, [3]*Riddet Institute, Massey University, Palmerston North, New Zealand*. Gastric dynamic *in vitro* models can help understand the effects of diet properties on gastric emptying, a key modulator of nutrient digestion kinetics. We assessed the capacity of the Human Gastric Simulator (HGS) to reflect the gastric emptying behaviour of eight diets differing in fibre properties, known to induce variation in gastric emptying *in vivo* in pigs. Diets contained either wheat bran, wood flour, sunflower hulls, fine-, medium- and coarse wheat straw, wheat bran plus low-methylated pectin, or wheat straw plus high-methylated pectin. Gastric emptying was assessed using tracers to follow liquids (Co- or Yb-EDTA), fine solids ($TiO_2$), and fibrous particles (Chromium mordanted fibres). Fine- wood flour, sunflower hulls and straw accelerated gastric acidification and reduced the half-emptying time ($T_{1/2}$) of fine solids (-17 to 157 min) and fibrous particles (-72 to 292 min; $P \leq 0.05$), reducing digesta phase separation (-5 to 108 min), compared with coarser wheat bran and straw. Pectin addition to coarse straw and bran delayed acidification, and reduced digesta phase separation (0 to -90 min), by increasing $T_{1/2}$ of liquids (+24 to 117 min; $P \leq 0.05$), and reducing $T_{1/2}$ of fibrous particles (-40 to 62 min, $P \leq 0.05$). Evaluation of *in vivo-in vitro* relationships showed similar ranking for gastric emptying of liquids and fibrous particles. At lower mean retention time (MRT) of liquids *in vivo*, the HGS could not discriminate differences among diets, and underestimated the retention of fine solids and fibrous particles at large MRT *in vivo*. Hence, digesta phase separation was underestimated *in vitro*. Overall, fine insoluble fibres accelerated gastric emptying and acidification compared with coarse insoluble fibres, while pectin addition reduced phase separation and delayed acidification, indicating the potential of the HGS to study the effects of diet properties on gastric emptying. Improving gastric sieving will benefit the accuracy of data generated by the HGS. **Key Words:** Human Gastric Simulator, Fibres, Gastric emptying

**165 Meta-analysis shows absence of a relation between digesta mean retention time and apparent ileal digestibility in growing pigs.** S. Dorado Montenegro*[1,2], W.J.J. Gerrits[1], and S. de Vries[1], [1]*Wageningen University & Research, Wageningen, Gelderland, The Netherlands*, [2]*University of Costa Rica, Montes de Oca, San José, Costa Rica*. A current dogma in digestive physiology is that prolonging the mean retention time (MRT) of digesta increases apparent ileal digestibility (AID) of nutrients by increasing exposure time to digestive enzymes. We examined the relation between MRT of digesta in the proximal gastrointestinal tract (GIT) and AID of dry matter (DM), starch (S), and nitrogen (N) in growing pigs. A meta-analysis was conducted using data from 185 pigs and 27 diets across six trials. The AID of DM was analyzed for the entire dataset. The AID of S and N was analyzed in a subset of 30 pigs and 8 diets from 2 trials. Stomach (ST) and small intestine

(SI) MRT were studied separately, relating solid digesta MRT with AID of DM and S, and liquid digesta MRT with AID of N. MRT was standardized for feed intake. Regression analyses were performed separately by trial to assess the effects of diet, MRT, and their interactions on AID. This allowed evaluating trial effects. Only 3 out of 27 diets from 2 trials showed an increase in AID of DM when solid digesta MRT in the SI increased, and these diets contained high levels of resistant S or beta-glucans. Subsequently, correlation analysis was performed using pooled data. No significant interactions were found between diet and MRT in the ST or SI ($P > 0.05$). Yet, dietary differences were observed in AID of DM when modifying MRT in the SI ($P = 0.009$) and in AID of S when modifying MRT in the ST ($P = 0.022$). In the ST, a negative relation ($r = -0.13$, $P = 0.087$) between solid digesta MRT and AID of DM was driven by 2 observations with an MRT exceeding 3 times the mean MRT of 189 minutes. This correlation disappeared after removing these observations. In the SI, MRT of solid digesta had no effect on AID of DM, and a positive correlation ($r = 0.14$, $P = 0.064$) was driven by 1 observation. No effects of MRT on AID of S or N were observed in any GIT segment. These results indicate that variation in digesta MRT in the proximal GIT does not explain changes in AID of DM, S, or N. **Key Words:** Digesta passage, Nutrient digestibility, Growing pigs

## Theme III Posters: Functional Ingredients and Utilization of Feed Resources for Improved Digestive Function and Nutrient Efficiency

**166 Acute enhanced liquid aspirin administration improves performance and intestinal function in nursery pigs.** S.C. Pearce* and B.J. Kerr, *USDA-ARS, Ames, IA, USA*. Aspirin (acetylsalicylic acid) is a nonsteroidal anti-inflammatory drug which is used for pain relief as well as an anti-inflammatory and fever reducer. However, it has been shown to cause negative effects to the gastrointestinal (GI) tract including lower GI bleeding, perforation and can impact the small intestine. Enhanced liquid aspirin (ELA) contains a combination of a salicylate compound, glycerin, triacetate, and saccharin which is more stable than aspirin alone and may reduce negative effects associated with aspirin on the GI tract, while exerting positive effects on inflammatory processes. The objective of this study was to evaluate oral ELA on healthy weaning pigs. 8 pigs per treatment were group housed and gavaged daily for 5 d with either saline (CON) or 2 mg/kg body weight ELA. After the 5-d dosing period, pigs were weighed and then euthanized for intestinal sample collection. Fresh ileum samples were collected for Ussing Chambers while additional samples were either snap frozen for cytokine and gene analysis or formalin fixed for histological analysis. ELA-treated pigs gained significantly more body weight compared to CON pigs (0.86 vs. 0.49 kg; $P < 0.05$). There were no differences in ileal permeability. There was tendency for an increase of 24% in ileal villus height in ELA pigs compared to CON ($P = 0.06$) and significant increases in relative protein expression of Claudins (CLDN) 3 and 7 ($P < 0.05$) in the ileum. Additionally, immune related genes DEFB1, IFNG, IL1B, and TLR2 were decreased in ELA pigs while REG3G was significantly increased ($P < 0.05$). Finally, stem signaling genes NOTCH1, WNT3A were also decreased in ELA-treated pigs compared to CON ($P < 0.05$). In conclusion, this data showed that ELA was well tolerated in a pig model and had no observable negative impacts. Additionally, ELA exerted several positive effects on body weight and intestinal function that would warrant follow up studies. **Key Words:** aspirin, barrier function, physiology

**167 Targeted dietary supplementation enhances growth in IUGR piglets: A study on probiotics, medium-chain triglycerides, and antioxidants.** R Ruggeri[1], G. Bee*[1], and B. Eichenberger[2], [1]*Agroscope, Posieux, Switzerland*, [2]*UFA, Herzogenbuchsee, Switzerland*. Improved sow prolificacy but also increased the number of piglets born with a low birth weight and those affected by intrauterine growth restriction (IUGR). These piglets typically show lower survival rates and stunted growth. This study aimed to evaluate the effects of a starter diet supplemented with probiotics, medium-chain triglycerides, and grape seed antioxidants on the growth of light and IUGR pigs. To determine the degree of IUGR, the brain-to-liver weight ratio of each piglet (n = 115) was estimated using 2 regression equations based on the piglets' body weight (BW), ear-to-ear distance, and abdominal area at birth. Additionally, pigs were categorized as either heavy (H = 56; BW = 9.6 ± 1.8 kg) or light (L = 59; BW = 6.2 ± 1.7 kg) based on their BW at weaning. The trial began one week after weaning and lasted 4 weeks. The control group (H-CON; n = 28; L-CON; n = 29) was fed a standard starter diet, while the treatment group (H-TRT; n = 28; L-TRT; n = 30) received a diet supplemented with probiotics, medium-chain triglycerides, and antioxidants. The individual BW of the pigs was recorded at weaning, day 1, 14, and 28. The average daily gain (ADG) was calculated from weaning to day 1, day 1 to 14, day 15 to 28, and day 1 to 28. The feed intake was measured per pen from day 1 to 28. Pigs from the treatment group showed higher BW ($P = 0.05$) and ADG ($P = 0.01$) by day 28 compared to CON pigs. However, L pigs and those with elevated brain-to-liver weight ratios, indicating IUGR, consistently grew slower ($P < 0.01$) regardless of the treatments. Throughout the trial, H pigs ingested more feed ($P < 0.01$) compared to the L group, regardless of the treatments. Among the L pigs, however, the treatment pens showed a greater feed intake ($P < 0.01$) than the control groups. These findings show that while dietary supplementation improves growth, more targeted strategies are needed to meet the specific needs of light and IUGR piglets and enhance production efficiency. **Key Words:** Weaning performance, production efficiency

**168 Evaluation of the complex additive in weaning challenged with *Escherichia coli*.** H Kim*, S Chang, D Song, K Jeon, J Yang, and J Cho, *Chungbuk national university, Cheong ju, Chungcheongbuk-do, Korea*. This study investigated the effects of complex additives in weaning pigs challenged with *Escherichia coli* (*E. coli*). The additive included essential oil (thymol and carvacrol), organic acids, vitamin E, and selenium. 24 LYD pigs (28 days old; 9.61 ± 0.81 kg) were used in a 17-d study, including a 3-d adaptation period. Pigs were assigned to four treatments: PC (basal diet), NC (PC + E. coli), T1 (NC + 0.05% additives), and T2 (NC + 0.10% additives). Challenged groups were orally administered 3 mL *E. coli* ($10^{1°}$ CFU/mL). PC reduced ($P < 0.05$) ADG, ADFI, villus height (VH), and tight junction ($P < 0.05$) while increasing crypt depth (CD) compared to NC. Tight junction proteins regulate the passage of harmful substances in the intestine. T1 had higher ($P < 0.05$) ADG and ADFI than NC at 0–7 d. At 28 d, the additive increased ($P < 0.05$) VH:CD ratio, claudin-1, and mucin-1 levels. T1 had a lower ($P < 0.05$) CD and higher ($P < 0.05$) zonula occludens-1 than NC. At 3 d, T1 and T2 had higher interferon-γ ($P < 0.05$), and T2 had higher ($P < 0.05$) interleukin-8 than PC. At 3 d, 16S rRNA sequencing of fecal samples revealed T2 differed ($P < 0.05$) in beta diversity from PC and T1. At the genus level, T2 had higher ($P < 0.05$) *Lactobacillus* and *Megasphaera*, while NC had lower *Phascolarctobacterium* and *Christensenellaceae* R-7 group levels than others. At 14 d, beta diversity differed ($P < 0.05$) between T2 and NC and PC. NC exhibited higher ($P < 0.05$) *Oscillosporaceae* and lower ($P < 0.05$) *Ruminococcus* than others, while PC showed lower ($P < 0.05$) *Parabacteroides* and higher ($P < 0.05$) *Prevotella* than others. Supplementation of complex additives enhances immunity during infection and improves growth performance and intestinal morphology by modulating gut microbiota. The optimal inclusion level of complex additives is considered to be 0.05%. **Key Words:** Weaning pig, *Escherichia coli*, complex addition

**169 Supplementing sows during lactation with fiber or a stimbiotic modulates fecal volatile fatty acid profile and calprotectin.** R. Self*[1], A. Waller[1], A.L. Petry[1], L. Merriman[2], P. Wilcock[2], S. Becker[2], R. Schmitt[3], H. Williams[3], J. Flohr[3], and R. Moreno[3], [1]*University of Missouri, Columbia, MO, USA*, [2]*AB Vista, Marlborough, Wiltshire, United Kingdom*, [3]*Seaboard Foods, Guymon, OK, USA*. This study aimed to assess the effects of fiber and stimbiotic supplementation during periparturition and lactation on fecal calprotectin and volatile fatty acids (VFAs). The mechanism of action of a stimbiotic activates fiber-degrading microbes, improving the capacity for fiber fermentation. In a randomized 2×2 factorial design, 860 sows (25% parity 1, 38% parity 2-3, 37% parity 4+) received either a corn (CON) or equal blend of wheat-middlings and soybean hulls (FIB) with or without a stimbiotic (+/-) top dress. Sows received 227g of top dress twice daily (0.454 kg/d) alongside their lactation diet (TDF=246 g/d, 372 g/d pre-farrow for CON and FIB, respectively), starting 1 day after entering farrowing stalls until weaning. Fresh fecal samples were collected for 120 sows at 5 days post-treatment application, and at weaning. Data were analyzed as linear mixed model with diet, stimbiotic, parity, and their interactions as fixed effects and farrowing room as a random effect. On day 5 post-supplementation, sows fed FIB+ had elevated butyrate levels (16.09 mM) relative to CON+ (11.69), CON- (11.71), and FIB- (13.2; Diet×Stimbiotic, $P = 0.016$). Similarly, on day 5, sows fed FIB had 10.7% greater fecal acetate concentration (Diet, $P = 0.04$). Fecal acetate concentrations at weaning were highest in CON+ sows (110.84 mM) compared to CON- (84.5), FIB- (86.92), and FIB+ (84.99; Diet×Stimbiotic, $P = 0.042$). There was a tendency for an increased concentration of butyrate in CON+ at weaning (Diet×Stimbiotic, $P = 0.09$). Irrespective of diet, stimbiotic supplementation increased propionate at weaning (Stimbiotic, $P = 0.014$). Fecal calprotectin levels tended to decrease in sows 5 days after stimbiotic supplementation (3.34 vs. 3.77 mg/g of protein; Stimbiotic, $P = 0.085$). Collectively, stimbiotic supplementation modifies VFA profiles during lactation indicating improved microbial fermentation and reduces intestinal stress biomarkers. These findings suggest potential benefits for sow gut health. **Key Words:** stimbiotic, fiber, sow

**170 Nutritional value of processed black soldier fly larvae for pigs.** A.J.M. Jansman* and P.G. van Wikselaar, *Wageningen Livestock Research, Wageningen University and Research, P.O. Box 338, 6700 AH Wageningen The Netherlands*. Ingredients based on insects such as black soldier fly (BSF) larvae (*Hermetia illucens L.*) are considered as novel feed ingredients for piglets. Prior to use as ingredient processing steps (heating, tricanter centrifugation and drying) are applied to separate the fat fraction and dry the product leaving BSF meal as protein source. The present study evaluated the ileal and faecal digestibility of differently processed BSF larvae (non-defatted, microwave dried (MD); heat treated (90°C, 4 h; DH) or enzyme (protease and lipase, 4 h) and heat treated (90°C, 4 h; DEH) during defatting prior to drying) in ileal canulated piglets using a change-over design with two experimental periods with twelve ileal canulated piglets (15-30 kg BW). In both periods three days of 12 h ileal digesta collection was preceded by two days of faeces collection. A basal diet and three experimental diets were used in which 20% of the basal diet was replaced by either batch of BSF. $TiO_2$ was used as marker. The difference method was applied to calculate the digestibility of BSF. Results were analysed by ANOVA and using the LSD test to compare treatments. Contents of crude protein were 445, 519 and 521 g/kg DM and of fat (EE) were 226, 142 and 139 g/kg DM for MD, DH, and DEH, respectively. Apparent ileal digestibility of N (CP) was 72.8, 68.1 and 68.0% ($P > 0.05$) and of amino acids did not differ among batches of BSF ($P > 0.05$). Apparent faecal fat digestibility for MD, DH, and DEH was 81.9, 86.7 and 91.6% ($P < 0.05$). The calculated net energy (NE) value based on faecal digestible nutrients (CVB, 2023) was 12.3, 11.1 and 10.8 MJ/kg DM for MD, DH and DEH, respectively. It can be concluded that BSF larvae are a potential protein source for piglets with an adequate nutritional value. Adapting processing conditions, including targeted enzyme processing in the defatting process, could further improve the nutritional value of BSF for piglets. **Key Words:** insect larvae, processing, digestibility

**171 Assessment of dietary Spirulina supplementation on growth, jejunal morphology, nutrient digestibility, and intestinal health-related genes in LPS-challenged weanling pigs.** E.O. Alagbe*[1], K.M. Ajuwon[1], H. Schulze[2], and O. Adeola[1], [1]*Department of Animal Sciences, Purdue University, West Lafayette, IN, USA*, [2]*Livalta, Peterborough, United Kingdom*. The intestinal health of weanling pigs is often compromised by harmful bacterial agents, leading to inflammation, and reduced intestinal integrity. Limited research exists on the effects of Spirulina (SP) as a bioactive feed additive on weanling pigs. Hence, this study aimed to investigate the effect of SP on the growth performance and intestinal health of pigs using a lipopolysaccharide (LPS; Sigma-Aldrich) challenge model. On experimental d 0, 128 pigs (7.4±0.7 kg, 28 d of age) were assigned to 1 of 4 groups in a 2 × 2 factorial arrangement of treatments with 2 levels each of SP (0 or 5 g SP/kg) and LPS (challenge or no-challenge). There were 8 replicate pens per treatment and 4 pigs per pen. On d 7, pigs were weighed, and pigs in the LPS challenge group were intraperitoneally injected with 100 µg/kg body weight LPS. The other pigs were injected with saline. On d 14, pigs were weighed; fecal samples were collected on d 13-15. On d 15, one pig per pen was reinjected with LPS or saline. After 4 h, injected pigs were euthanized for sample collection. Data were analyzed using PROC MIXED of SAS and evaluated for main effects and interactions. There was no SP effect on growth performance. However, the LPS challenge reduced ($P < 0.05$) the body weight and gain-to-feed ratio of pigs. Dietary SP improved ($P < 0.05$) the apparent total tract digestibility (ATTD) of dry matter regardless of the LPS challenge. There was an SP × LPS interaction ($P < 0.05$) on the ATTD of energy, jejunal villus height, and villus area. Dietary SP increased ($P < 0.05$) the mRNA expression of jejunal zonula occludens-1. An SP × LPS interaction ($P < 0.05$) showed that LPS-challenged pigs fed an SP-supplemented diet had the highest jejunal superoxide dismutase 2 mRNA expression relative to pigs in the other groups. In summary, dietary Spirulina enhanced antioxidant activity in pigs during an LPS challenge and improved tight junction integrity and nutrient digestibility, irrespective of the LPS challenge. **Key Words:** intestinal health, lipopolysaccharide, spirulina

**172 Hydrolyzed yeast a valuable component in ZnO replacement strategies for pigs post-weaning.** H Schulze*[1] and S Kaczmarek[2], [1]*Livalta, Peterborough, UK*, [2]*University of Life Sciences, Poznan, Poland*. The use of therapeutic levels of zinc oxide (ZnO) is increasingly regulated in many regions. Yeast products, due to their gut modifying properties can be a viable alternative in ZnO replacement strategies. This study aimed to investigate the effect of hydrolyzed yeast (*Saccharomyces cerevisiae*; HY) on growth performance parameters and well-being of weaned pigs fed diets without therapeutic levels of ZnO. The study included five dietary treatments, with (PC) or without therapeutic ZnO addition. The treatments without therapeutic ZnO were supplemented with 0 (NC), 1, 2 or 4 kg/ton HY. Mixed cereal pelleted starter (d 1-21) and weaner (d 22 – 42) diets were fed ad libitum. Ten individually housed pigs of initial average 7.5 kg body weight and 28d of age were used per treatment. Body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR) were measured per pig for each period and overall. Piglet faeces consistency (diarrhea score) was recorded daily during d 1-14, 21-28, 35 and 42. Faeces was collected twice daily per pen during d 10-14 and 32-36, frozen, and pooled per treatment and period before analysis. Total tract nutrient digestibility was calculated using the TiO marker method. The experiment was conducted as a completely randomized design and results analyzed by ANOVA procedure in R using the “Agricolae” package. The removal of therapeutic ZnO (PC vs NC) resulted in reduced ($P < 0.001$) BWG and FI during each period and overall, while FCR was increased ($P < 0.001$) during the starter period and ($P < 0.01$) overall. Starting from an inclusion of 2 kg/ton, HY restored BWG and overall FCR to those observed in PC. Pigs of NC had highest diarrhoea scores ($P < 0.05$ at d 4, 8, 11, 14, 24, 25, 35 and 42). Therapeutic ZnO (PC), or high doses of HY improved faeces consistency to the scores observed in the PC treatment. In summary, HY addition can be a viable alternative in ZnO replacement strategies helping to maintain well-being and performances of pigs post-weaning. **Key Words:** ZnO replacement; hydrolyzed yeast; growth performance; diarrhoea

**173 Nutrient and energy digestibility of sorghum protein concentrate in growing pigs.** J. A. L. Barbosa[1], H. Moreira Júnior[1], A. Gorrosterrazú[1], J. L. Brito[1], C. E. M. Bertanha[1], S. S. S. Sousa[1], M. L. P. Tse[2], and U. S. Ruiz*[1], [1]*University of São Paulo, Piracicaba, São Paulo, Brazil*, [2]*São Paulo State University, Botucatu, São Paulo, Brazil*. Sorghum Protein Concentrate (SPC) is a feed ingredient obtained after starch removal from sorghum in a wet milling process. SPC can be used in swine feeding, but there is no information about the digestible nutrient and energy profile of this feedstuff. Thus, the aim of this study was to evaluate the standardized ileal digestibility (SID) of amino acids (AA) and crude protein (CP), and the apparent total tract digestibility (ATTD) of dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF), ether extract (EE), and gross energy (GE), digestible energy (DE), metabolizable energy (ME), and estimated net energy (NE) in growing pigs. Twenty animals (25.34 ± 2.16 kg) were fed a nitrogen-free diet (NF) or a diet with 40% SPC, as the only source of nitrogen, replacing starch, sugar, cellulose and oil of the NF, and both diets contained 0.5% titanium dioxide as an index compound. The animals were housed in individual pens, had free access to water, and were fed at 2.8 x maintenance DE for 11 days (9 for adaptation and 2 for partial feces collection). On day 11, pigs were euthanized for ileal digesta collection. A randomized block design was used with 10 replicates and 1 animal per experimental unit. The SPC contained (as is basis) 93.58% dry matter, 53.62% CP, 5751 kcal/kg GE, 16.56% EE, 11.30% starch, 2.62% NDF and 1.59% ADF. The AA profile resembled that of sorghum, with high contents of Glu (11.61%), Leu (6.34%), Asp (5.25%), Ala (4.36%), Val (3.02%), Thr (2.81), Ile (2.58%), Phe (2.57%), and low levels of Lys (1.00%), Met (0.65%), and Trp (0.42%). The ATTD of DM, OM, CP, EE, and GE ranged from 76-82%, and the ATTD of NDF and ADF were 72% and 52%, respectively. The DE, ME, and NE contents (as is basis) were 4428, 4064, and 3134 kcal/kg. The SID of total AA was 85.44%, ranging from 73.72% to 95.00%. In conclusion, SPC has high nutritional value for swine due to the high digestibility of its components. **Key Words:** Coproducts cereal

**174 Meta-analysis of Exogenous Fibre Enzymes in Modulating the Apparent and True Total Tract Crude Fat Digestibility and the Fecal Endogenous Losses of Crude Fat in Weanling Pigs.** Laurence Cheng*, Jiali Chen, Min Wang, and Ming Fan, *Department of Animal Biosciences, University of Guelph, Guelph, ON Canada N1G 2W1*. Dietary fiber has been recognized as an anti-nutritive factor, limiting dietary nutrient digestibility and profit margin in pig production. For over 2 decades, exogenous fibre enzymes have been used as feed supplements in swine industry; however, their efficacy in modulating crude fat digestibility in weanling pigs remains inconsistent in the literature. This study aimed to evaluate if exogenous fiber enzymes modulate crude fat digestibility and fecal endogenous fat losses in weanling pigs. We carried out a literature review and meta-analyses of studies published during 2004-2023 and examined the effects of exogenous fiber enzyme supplementation on the crude fat digestibility in weanling pigs fed commercial diets. Sources of studies were treated as a randomized block factor; data normality was checked by the Shapiro-Wilk test and all statistical analyses were carried out by SAS. There were no differences ($P > 0.05$) in the least square means (LSMEAN ± SEM) of the initial BW (7.6 ± 0.5 vs. 7.6 ± 0.7 kg; n=17 vs. 13), dietary NDF content (11.0 ± 1.3 vs. 11.0 ± 1.4%; n=13 vs.11) and dietary crude fat content (5.4 ± 0.5 vs. 5.5 ± 0.5%; n=19 vs. 12) between the non-fibre enzyme (Non-enzyme) and the exogenous fibre enzyme-supplemented (Fibre enzyme) groups. Kruskal-Wallis non-parametric contrast showed no difference ($P > 0.05$) in the apparent total tract crude fat digestibility (LSMEAN±SEM, 73.1 ± 3.8 vs. 74.3 ± 4.0%; n=32 vs. 16) between the Non-enzyme and the Fibre enzyme groups. There was no difference ($P > 0.05$) in the total tract true crude fat digestibility (parameter estimate ± SE, 90.3 ± 9.6 vs. 104.8 ± 15.6%; n=13 vs. 6). Fecal endogenous crude fat losses were obtained via simple linear regression analysis and showed difference (LSMEAN±SEM, 7.6 ± 2.1 vs. 16.1 ± 2.3 g/kg dry matter intake; $P < 0.0001$; n=13 vs. 6) between the Non-enzyme and the Fibre enzyme groups. Our results suggest that the current commercial exogenous fiber enzymes do not positively affect the digestive utilization of dietary crude fat in weanling pigs. **Key Words:** Exogenous fibre enzymes; crude fat; digestibility

**175 Evaluation of the Impact of Flavoring Compounds on the Performance of Sows and their Progenies.** H Tran*[1], M Puyalto[2], L Pandolfini[2], J José Mallo[2], and B de Rodas[1], [1]*Purina Animal Nutrition, Gray Summit, MO, USA*, [2]*Norel Animal Nutrition, Pasadena, TX, USA*. Two experiments (EXP) were conducted to evaluate the effects of flavoring compounds (Spiced, (PA) and Raspberry Peach (FL), Norel Animal Nutrition) on the performance of lactating sows and their progenies. In EXP. 1, 38 sows (PIC, average parity = 3.2) and their litters were used. At d 109 of gestation, sows were blocked by parity and farrowing date, and assigned to one of two dietary treatments: Control (Control; n = 19) and Control + PA at 0.0375% (PA; n = 19). Sows received their respective diets from d 109 of gestation to weaning. Data indicate that sows fed PA returned to estrus sooner (-0.5 day; $P < 0.05$) than the control. This improved outcome could be attributed to better feed intake of sows fed PA (+0.23 kg/day; $P > 0.10$). In EXP. 2, 504 weaned pigs (19 d of age; initial BW = 6.7 kg) born to the sows from EXP. 1 were used to evaluate the effects of feeding PA and FL on the performance of nursery pigs. At weaning, pigs were blocked by initial BW, gender, and sow treatment and allotted to pens, which were assigned to dietary treatments (12 pens/TRT; 10 to 11 pigs/pen) in a 2×2 factorial design, based on sow feeding (Control or PA) and nursery feeding (Control or PA in Phase 1, followed by FL in Phase 2 and 3). Pigs and feeders were weighed on d 0, 8, 15, 22, and 42 for ADG, ADFI, and F:G calculations. During d 0-8, feeding PA had no significant effect on ADG, ADFI, or F:G in the nursery. During d 22-42 and d 0-42, nursery pigs fed PA and FL tended ($P$, interaction < 0.10) to have better feed efficiency compared to pigs that did not receive PA and FL, regardless of the sow-feeding. In addition, feeding PA and FL to nursery pigs improved pen fecal firmness during d 0-22 ($P$, nursery effect < 0.01). Data from this study suggests that feeding PA and FL in the nursery can improve feed efficiency and fecal firmness compared to the control pigs. **Key Words:** flavoring compounds, sow, and nursery pigs

**176 Meta-analysis of the Digestive Utilization of Dietary Copper as affected by Exogenous Phytase Supplementation in Weanling Pigs.** Mingli Xu, Jiali Chen, Laurence Cheng, Min Wang, and Ming Fan*, *Department of Animal Biosciences, University of Guelph, Guelph, ON N1G 2W1*. Exogenous phytases have been widely used as feed enzymes in the swine industry for more than 4 decades. However, there are inconsistent literature reports of efficacy of the exogenous phytases in affecting the digestive utilization of dietary copper (Cu) in weanling pigs. A meta-analysis was conducted to determine the effect of dietary exogenous phytases (500–1000 FTU/kg) on the digestive utilization of dietary Cu in the weanling pigs. This Meta-analysis study included work published between 1992 and 2023 on weanling pigs fed dietary Cu levels within Cu requirements without or including exogenous phytases. Sources of studies were treated as a randomized block factor; data normality was checked by the Shapiro-Wilk test and all statistical analyses were carried out by SAS. The Kruskal-Wallis contrast showed no difference in the apparent fecal Cu digestibility (16.8±5.2 vs. 22.0±6.4%; $P$ = 0.53; n = 23 vs. 10) between the non-phytase and the phytase groups. There was an exponential relationship [Y = exp(0.175±0.059x); $R^2$ = 0.36; $P$ < 0.05; n=23] between the contents (mg/kg dry matter diet intake, DMI) of the apparent fecal digestible Cu and the total dietary Cu in the non-phytase group, suggesting that the true fecal Cu digestibility was affected by dietary Cu levels. There were linear relationships (y = 0.339±0.119x; $r^2$ = 0.28, $P$ < 0.05; n = 23 for the non-phytase group vs. y = 0.275±0.110x; $r^2$ = 0.44; $P$ < 0.05; n = 10 for the phytase group] between the contents (mg/kg DMI) of the apparent fecal digestible Cu and the total dietary Cu, thus estimating the true fecal Cu digestibility values between the two groups. The Kruskal-Wallis contrast showed differences in the fecal endogenous Cu outputs (2.331±0.759 vs. 0.656±0.967 mg/kg DMI; n = 23 vs. 10; $P$ = 0.0052) between the non-phytase and the phytase groups. Thus, our meta-analysis results indicate that exogenous phytases do not improve the apparent and true fecal Cu digestibility but significantly reduce the fecal endogenous Cu loss in the weanling pigs. **Key Words:** phytase; copper; pig

**177 Technical impact of a synergistic blend of organic acids and phytogenic compounds fed in late finishing diets: Meta-analysis using global data.** M. De Vos[1], S. Crowder*[2], R. Van Erp[3], D. Vergaelen[1], B. De Rodas[4], and R. D'Inca[1], [1]*Agrifirm, Drongen, Belgium*, [2]*Fortiva, Arden Hills, MN, USA*, [3]*Agrifirm, Apeldoorn, The Netherlands*, [4]*Purina Animal Nutrition, Arden Hills, MN, USA*. Feed represents a major cost in pig production, especially during the late finishing phase when feed efficiency is the lowest. Therefore, production chains continually look for improvements in genetics, housing and feeding practices. The latter includes feeding various functional ingredients to improve technical parameters. However, the different rearing practices across the world can lead to variable technical outcomes. This study evaluated the impact of a synergistic blend of organic acids and phytogenic compounds (AMB) on performance of finishing pigs. To properly estimate the efficacy of AMB, 20 animal experiments, with a total of 272 replicates/group, performed in North America, Europe and other regions were included. A random-effects meta-analysis was conducted using the rma() function (*metafor* package of R). The standardized mean difference (SMD) was calculated for each study prior to the analysis and used along with their corresponding variances as input for the meta-analysis. To facilitate interpretation, model estimates were subsequently transformed back to the original scale. Between-study heterogeneity was modeled, and the between-study variance ($\tau^2$) was estimated using the restricted maximum likelihood (REML) method. To assess the effect of geographical location, region was included as fixed effect into the model. This meta-analysis using global data demonstrates a significant improvement in late finishing performance when supplementing AMB. The synergistic blend of organic acids and phytogenic compounds improved ADG by +30.9 g/d ($P$ < 0.01), FCR by -0.09 ($P$ < 0.01) and had no impact on ADFI ($P$ > 0.05). No difference between regions was observed ($P$ > 0.05), confirming a global efficacy. However, animal experiments performed in North America seem to be numerically more efficient. In conclusion, adequately designed functional feed ingredients can improve performance of pigs in different rearing or nutritional contexts. **Key Words:** Finishing, Efficiency, Functional

**178 Metabolomic profiling of plasma responses to vitamin D and C supplementation in a postweaning pig disease model.** M. V. Curtasu*[1,2], D. Bueno Dalto[3], C. A. Gagnon[4], L. Cloutier[5], F. Guay[2], and M. P. Létourneau-Montminy[2], [1]*Aarhus University, Faculty of Technical Sciences, Department of Animal and Veterinary Sciences Campus Viborg, Blichers Alle 20, 8830 Tjele, Denmark,* [2]*Laval University, Faculty of Agriculture and Food Sciences, Department of Animal Sciences, 2425 rue de l'Agriculture, Québec, G1V 0A6, Canada,* [3]*Sherbrooke Research and Development Centre, Agriculture and Agri-Food Canada, Sherbrooke, Québec J1M 0C8, Canada,* [4]*Swine and Poultry Infectious Diseases Research Center, Faculté de Médecine Vétérinaire, Montreal University, 3200 rue Sicotte, Saint-Hyacinthe, Québec, J2S 2M2, Canada,* [5]*Centre de développement du porc du Québec (CDPQ), 815 Rte Marie-Victorin, Lévis, Québec G7A 3S6, Canada*. The objectives of this study were to evaluate vitamin supplementation as a holistic strategy to improve health status under a natural disease challenge with Porcine Reproductive and Respiratory Syndrome virus (PRRSv) and to study the effects on plasma metabolome profiles. Nine batches of weaned piglets (21 days old, n=540) were used to test the effects of functional vitamins: 25(OH)D3 and vitamin C. Three diets were formulated: VitD (0.05 mg 25(OH)D3), VitDC (0.05 mg 25(OH)D3 plus 500 mg vitamin c) and CON (no 25(OH)D3/Vitamin C). Postweaning piglets were fed the experimental diets for 19 days before exposure to the challenge (day 0) and continued for 28 days post-challenge. All batches were confirmed PRRS-positive post-exposure with a type 2 strain (L1H subtype based on ORF5 classification). Plasma was collected from 4 pigs/pen at days 0, 14, and 28 post-challenge for untargeted metabolomics profiling using high-resolution mass spectrometry. Health score data were analyzed with linear mixed-effects and multivariate models for metabolomics. Piglets on the VitD diet showed improved health scores between days 14–21 ($P = 0.042$). Partial least square regression analysis revealed a better separation of VitDC metabolite profiles from CON compared to VitD. Methylglutamine and pantothenic acid were found at lower intensities in plasma from pigs on VitDC diet ($P < 0.05$) at day 14 and 28 but not day 0. Several bile acid metabolites were higher in VitD pigs at day 0, 14, 28 compared to CON indicating a previous modulation due to the 19 days supplementation prior to PRRSv exposure. Moreover, metabolites from the purine degradation (guanine, xanthine, deoxyuridine) were lower in intensity in the plasma of VitD pigs at day 0, 14, 28 ($P < 0.05$). In conclusion, functional vitamin D supplementation indicated an improved health status and highlighted modifications of specific pathways within the plasma metabolome, although the underlying mechanisms warrant further investigation. **Key Words:** metabolomics, functional ingredients, vitamins

**179 Olive bioactives increase the resilience of immune challenged weaned piglets similarly to high doses of ZnO.** S. López-Vergé*[1], J.J. Pastor[1], E. Otto-Tice[2], and G. Tedo[1], [1]*Innovation division, Lucta S.A., UAB Research Park, Campus UAB, Cerdanyola del Vallès, Barcelona, Spain,* [2]*Lucta US, Industrial Avenue Mahwah, New Jersey*. Young piglets must face different challenges (weaning, transport, environmental changes, etc) that may impair their gut health, compromising their welfare and performance. The ban of ZnO forced the swine sector to look for alternatives to increase piglets' resilience. In this sense, olives are known to be a great source of bioactives with proven antioxidant and anti-inflammatory activities, among others. Therefore, the present study aimed to evaluate the potential benefits of olive bioactives (OBE) in acute immune-challenged piglets. Forty-eight mixed sexes piglets Pietrain x (Landrace x Large White) were used. At weaning (24d ± 2.9d), piglets were randomly distributed in 4 treatments (6 piglets/pen) by initial BW (T1: Control; T2: LPS; T3: LPS+ZnO, 3000 ppm; T4: LPS+OBE, 2500 ppm). Piglets were intraperitoneally injected with either a solution containing 50µg/kg BW of LPS (Serotype O55:B5) or the same amount of 0.9% NaCl (saline) solution on day 21 post-weaning (PW). Blood and tissue samples were collected 4h after the LPS injection. No differences in performance were observed among treatments. The ZnO and OBE groups tended to increase the concentration of PigMAP in plasma (P = 0.09) compared to T2 to similar levels than the control group (T1) after the LPS challenge. In liver, ZnO and OBE groups upregulated the expression of *IL10* ($P < 0.05$), suggesting a better anti-inflammatory status after the LPS challenge. Finally, in Jejunum both ZnO and OBE groups tended to improve the barrier function by regulating the expression of *ZO-1* (P = 0.06) and *CLAUDIN-1* ($P = 0.06$). In summary, these

results suggest that the administration of OBE in weaned piglets contributed to control the side effects of an LPS challenge by improving their anti-inflammatory status and their gut integrity. This may be a good strategy to help animal producers in the development of new natural strategies to effectively increase piglet resilience in the absence of therapeutical doses of ZnO. **Key Words:** Olive bioactives, weaning, resilience

**180 The glucose and nitrogen release characteristics of multiple feed ingredients for pigs.** M. Huang*, Y. Xu, J. Li, Y. Cao, W. Huo, Z. Zhang, J. Zhao, D. Han, D. Li, and J. Wang, *College of Animal Science and Technology, China Agricultural University, Beijing, China*. The objective of this study was to investigate the release kinetics of glucose, soluble nitrogen, and amino acids from pig feed ingredients during *in vitro* digestion, providing insights into the optimal utilization of feed resources. Using a two-step enzymatic process, we measured the release of glucose, nitrogen, and total amino acids from 22 feed ingredients (such as corn, soybean meal, etc.) at various time points during simulated gastric and intestinal phases. Deng model was used for estimating soluble nitrogen release rate in the gastric phase. For the other estimations, the first-order kinetic model was applied. Kruskal-Wallis test was used for statistical analyses. The study revealed that glucose release rate varied among ingredients ($P < 0.01$), ranging from 0.0103 to 0.0754 %/min during the intestinal phase. Soluble nitrogen release rates in protein feeds differed significantly ($P < 0.001$), both in gastric (0.21 to 27.68 %×$10^{-4}$/min) and intestinal (40.00 to 929.86 %×$10^{-4}$/min) phase. Though total amino acid release rates fluctuated (0.0045 to 0.0127 mmol/g CP /min), there was no significant difference among ingredients. Furthermore, different feed ingredients exhibited distinct release patterns at various phase. For example, brown rice, wheat, and broken rice released glucose quickly in the early stages of the simulated intestinal phase. During the mid-intestinal phase, the release rates of glucose from some feeds, like corn and sorghum, accelerated, while others, like rice bran, slowed down. Protein feed ingredients like fish meal and rice protein released nitrogen quickly in the initial gastric digestion phase, but slowed down in the subsequent intestinal digestion phase. Our study highlights varying release rates of nutrients among feed ingredients and nutrient release differed widely across digestion phases. Future research should focus on formulating diets that consider these release kinetics to optimize pig growth performance and resource utilization. **Key Words:** glucose release kinetics, nitrogen release kinetics, feed resources utilization

**181 Effects of a sulfonating feed additive on the absorption and metabolism of deoxynivalenol in growing pigs.** M. L. McGhee*[1], R. J. Faris[1], D. W. Giesting[1], P. Pillai[1], C. M. Crincoli[1], W. Mosher[2], and C. Chen[2], [1]*Cargill, Inc., Wayzata, Minnesota, USA*, [2]*University of Minnesota, St. Paul, Minnesota, USA*. This study investigated the effects of a sodium metabisulfite (SM)-based feed additive on the disposition of deoxynivalenol (DON) in growing pigs by quantifying DON and its metabolites [DON-sulfonates (DONS-1, DONS-2, and DONS-3) and de-epoxy-DON (DOM)] in digesta, fecal, and urinary samples. Individually-housed pigs (n=48, 25.7 ± 1.6 kg) were allotted to 4 dietary treatments formulated to include approximately 1 ppm or 2 ppm DON, with or without SM, and acclimatized to diets for 17 d. Pigs were placed in metabolism crates on d 14 of study, followed by 3-d adaptation and 3-d fecal and urine collection. On d 20, pigs were euthanized. Digesta samples from the stomach, jejunum, ileum, and cecum were collected. The concentrations of DON and its metabolites were determined by liquid chromatography-mass spectrometry. Data were analyzed using a mixed model with fixed effects of DON, SM, and their interaction, and random effects of block (6 pigs/block), group (12 pigs/group), and replicate. Inclusion of SM resulted in extensive conversion of DON to DONS in feed—primarily DONS-2. Higher DON diets resulted in greater ($P < 0.05$) DON concentrations in digesta (stomach, jejunum, and ileum), feces, and urine. Inclusion of SM reduced ($P < 0.05$) DON concentration in the stomach, jejunum, ileum, and urine. Likewise, SM increased ($P < 0.05$) concentrations of DONS in digesta at all locations and feces. Interactions ($P < 0.05$) were observed due to greater DONS formation in the higher DON treatment level. Relative proportions of DONS-2 and DONS-3 shifted from higher DONS-3 in anterior samples to more

DONS-2 in posterior and fecal samples. Reduced urinary DON output ($P < 0.05$) and increased urinary DOM output ($P < 0.05$) was observed with SM. To conclude, SM reduced DON and increased DONS in feed and throughout the gastrointestinal tract, interconversion between DONS forms were observed in the gut, and excretion of DON metabolites suggests less DON absorption occurred when SM was fed. **Key Words:** deoxynivalenol, mass balance, mycotoxin

**182 Feeding a synergistic blend of organic acids and phytogenic compounds improves growth performance of finishing pigs.** M. De Vos*, S. Tanghe, M. Intven, K. Lannoo, and R. D'Inca, *Agrifirm, Drongen, Belgium*. During pig rearing, feed constitutes a major economic and ecological expense, particularly during the finishing phase when feed efficiency of animals is at its lowest. The present study was conducted to evaluate the effect of a synergistic blend of organic acids and phytogenic compounds on growth performance in finishing pigs. The product (AMB) was designed to improve health and microflora of the intestine while reducing oxidative stress and inflammation, therefore allowing for a better feed utilization and resulting in a more efficient growth. A total of 1,000 crossbred grower pigs, weighing 38.1 ± 3.2 kg were distributed to 40 straw-based pens (25/pen), split by sex. The feeding program was split in 3-phases (d0-28, d28-42 and d42-83). Experimental diets were fed *ad libitum* during the finisher phase (d42-d83) and were either a control diet or a test diet supplemented on-top with AMB (1kg/T). Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. The data were analyzed by mixed models with pen as experimental unit and body weight at d42 as covariate. Mortality data were analyzed using a Kaplan-Meier survival analysis. Differences were considered significant at $P < 0.05$ and trends at $P < 0.1$. Results showed that the supplementation of synergistic blend of organic acids and phytogenic compounds significantly improved ADG (+8.2%; $P < 0.01$) and decreased FCR (-4.7%; $P = 0.04$). No differences in ADFI nor mortality could be observed ($P > 0.05$). To conclude, this study showed that the supplementation of a specifically-designed blend of functional compounds during the last 41 days prior to slaughter enhances growth performance and contributes to sustainability in pig production through improved feed efficiency. **Key Words:** Functional ingredients, efficiency, performance

**183 Effects of dietary supplementation with olive oil wastewater extract on growth performance and fecal microbiota of weaning pigs.** G. Battacone*[1], F Correa[2], M. R. Mellino[1], D. Luise[2], G. Bee[3], and P. Trevisi[2], [1]*Department of Agricultural Sciences, University of Sassari, 07100 Sassari, Italy*, [2]*Department of Agricultural and Food Sciences, University of Bologna, 40127, Bologna, Italy*, [3]*Agroscope, 1725 Posieux, Switzerland*. Weaning in piglets is characterized by intestinal immaturity and oxidative stress, increasing vulnerability to post-weaning diarrhea (PWD) caused by enterotoxigenic *E. coli* (ETEC). Polyphenols, with antioxidative, antimicrobial, and gut eubiosis-promoting effects, show potential for supporting gut health during this period. This study investigated the impact of olive oil wastewater extract (OOW), a polyphenol-rich byproduct, on growth performance and fecal microbiota of ETEC genetic resistant or -susceptible healthy piglets during weaning. Forty-eight piglets (25 ± 1 days old) were randomly assigned to three dietary groups balanced for litter, sex, and ETEC F4 genetic susceptibility, based on the MUC4 gene polymorphism: CON (control diet), OOW1 (control + 0.05% OOW), and OOW2 (control + 2% OOW). Each group included 9 resistant and 7 susceptible pigs. Data were analyzed using a linear mixed model using treatment, genotype, run and day as fixed effects, and pig nested within pen as random effect. Average daily feed intake and average daily gain were higher in OOW1 compared to CON between d8 and d19 ($P = 0.02$, $P = 0.06$). Fecal scores in OOW2 were significantly lower than CON at d7 and d10 post-weaning ($P < 0.05$). OOW1 and OOW2 had a higher bacterial alpha diversity on d10 and d19 ($P = 0.009$). Microbial composition was influenced by the diet ($P = 0.002$), sampling day ($P = 0.001$), and genotype ($P = 0.01$). OOW reduced the abundance of *Escherichia-Shigella* (OOW1 vs CON, $P$-adj < 0.001, Log2 FC = -4.22; OOW2 vs CON, $P$-adj = 0.01, Log2 FC = -2.80) and *Campylobacter* (OOW1 vs CON, P-adj < 0.001, Log2 FC = -4.85; OOW2 vs CON, $P$-adj < 0.001, Log2 FC = -4.71). In conclusion, OOW supplementation

improved ADG and fecal scores while reducing pathogenic bacteria linked to PWD in piglets. The OOW have beneficial effect on the gut physiology of weaned pigs. Moreover, influence of genotype on microbes underscores the importance of considering host genetics in nutrition studies. **Key Words:** co-products, ETEC, polyphenols

**184 The effect of a *Bacillus amyloquefaciens* multi-strain probiotics on growth performance in weaned pigs: a meta-analysis of 4 trials.** Deepak E. Velayudhan*[1], Chong Shen[2], and Ester Vinyeta[1], [1]*Danisco Animal Nutrition & Health (IFF), Oegstgeest, The Netherlands*, [2]*IFF Nutrition & Biosciences, Brabrand, Denmark*. Early colonization of the gut with potentially beneficial and diverse gut microbes, can influence the maintenance of intestinal homeostasis and prevent gut dysbiosis in weaned pigs resulting in better performance. This meta-analysis evaluated the effect of a *Bacillus* sp. multi-strain probiotic fed continuously to pigs from day 1 until day 42 post-weaning on growth performance. The database was constructed using 4 studies [Landrace × Yorkshire × Duroc or PIC 410 × Libra Hyporc or Large White × Landrace × Duroc or Large White × Duroc breeds; average initial body weight (BW) of 6.9 ± 1.3 kg], contributing 680 data points. Pigs were fed corn or corn-wheat or rice-corn-based diets with soybean meal, fish meal (1 study), whey, wheat bran, or wheat middlings. Diets were fed as mash in 2 phases (1-14 and 14-42 days). All studies had 1) a control diet formulated to be nutrient-adequate, containing no added antibiotic growth promoters and pharmacological level of Zn; 2) the control diet supplemented with a 3-strain *Bacillus amyloliquefaciens* probiotics providing $3.0 \times 10^8$ cfu/kg of feed. Pigs were weighed individually, feed intake was recorded per pen at the end of each phase, and average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. Data were analyzed using Fit Model platform of JMP 16.1 with the study as a random effect. The BW of pigs was increased (9.9 vs 10.1 and 24.0 vs 24.5 kg; $P < 0.05$) on days 14 and 42 with supplementation of probiotics, respectively. The ADG was improved ($P < 0.05$) by 7.6, 2.4 and 3.0% and the FCR was reduced by 8.0, 3.7 and 3.2% for periods 1-14, 14-42 and 1-42, respectively. Diarrhea scores were observed to be either normal (score 1; normal feces) in both treatments or improved in probiotics-supplemented diets. compared to control. In conclusion, supplementing a multi-strain probiotics to commercially relevant diets improved the BW gain and feed efficiency in weanling pigs. **Key Words:** meta-analysis, *Bacillus* sp. multi-strain probiotics, pigs

**185 Comparison of alternative indicators to assess nutrient digestibility in pigs.** R. G. Lizardo*, J. G. Vazquez, and J. L. N. Ramos, *IRTA, Constanti, Tarragona, Spain*. Nutrient digestibility (DIG) is usually done by total collection (TC) of feces in metabolism crates or by the indicator method using indigestible markers like titanium dioxide ($TiO_2$) or acid-insoluble ash (AIA). However, the EU banned the use of $TiO_2$, the use of metabolism crates is questioned and results with AIA are highly variable. Consequently, alternative markers such as trivalent metals (yttrium (Y), ytterbium (Yb)) are needed. In trial 1, eighty 12 kg pigs were individually housed in metabolism crates for 5-day TC after 7-day adaptation. Piglets were offered wheat, barley, rice by-products and soybean meal-based diets containing 100 mg Y and Yb, and 10 g celite per kg of feed, respectively, in two meals per day. In trial 2, 25 kg pigs were offered *ad libitum* wheat, barley, maize and soybean meal-based diets containing the same amount of 100 mg of Y and Yb, and 5 g of $TiO_2$ per kg of feed for 14 days. At the end, pigs were slaughtered, and a grab faecal sample was collected from the rectum. All the samples were freeze-dried and ground before lab analyses. In trial 1, DIG values of DM, OM, CP, energy, and ash showed less than 1 percentage point difference for Y and Yb and correlate perfectly ($r > 0.992$); values obtained with AIA were 3-5 percentage points lower than with Y/Yb, and were only moderately correlated ($0.563 < r < 0.748$); no correlation was observed for ash ($r = 0.07$). In trial 2, DIG values of DM, OM, CP, energy, ash, Ca, P and Mg were similar and correlate perfectly between Y and Yb ($r > 0.997$); values obtained using $TiO_2$ were only less than 1 percentage point than with Y/Yb, and were highly correlated ($0.816 < r < 0.988$). Differences related with dietary treatments tested were equally observed with Y or Yb and $TiO_2$. Based on these results, it can be concluded that either Y or Yb give reliable results, much better than those observed with AIA, and they can perfectly replace $TiO_2$ as

indigestible marker to estimate nutrient digestibility. **Key Words:** Digestibility, indicators, methods

## Theme IV Posters: Development of Digestive and Absorptive Capacity in the Neonate and Impact of Weaning on Intestinal Function

**186 Gut health in piglets in antibiotic-free diets.** J.R. Pluske*, *Faculty of Science, The University of Melbourne, Parkville, Australia*. Antibiotics and mineral compounds [zinc oxide (ZnO), copper sulphate; collectively, antimicrobials] have traditionally been capable strategies to assist with the post-weaning transition and in preventing, controlling and (or) treating certain enteric diseases. These compounds have generally improved health and performance of post-weaned piglets, lowered mortality, and improved the cost of production. However, antimicrobial resistance, risks to the environment e.g., feeding ZnO, and changing market expectations and sentiments have curtailed, or banned, some antimicrobials in diets in a growing number of jurisdictions. As such, the adoption of "antibiotic-free" (ABF) production (sometimes interchanged with Raised Without Antibiotics – RWA) has attracted attention in some segments of the industry, although a precise definition of ABF is not necessarily without confusion and misunderstanding. In the nursery, the adoption of ABF production can be fraught, especially if medicinal levels of ZnO also cannot be used. The post-weaning transition can cause significant dysbiosis to the piglet's immature gastrointestinal tract (GIT), negatively impacting this organ's structure and function ('gut health'), and the absence of antimicrobial compounds can exacerbate the issue. Producers can implement best practices in e.g., biosecurity, health care, welfare, weaning management, genetics, staff training, feed handling and nutrition (e.g., feed additives, fibrous by-products), to the degree that is possible from a practical and economical perspective, to rear piglets in an ABF environment. From a nutritional perspective alone, there is a wide array of feeding and ingredient/additive strategies that have been commercially implemented that adhere to ABF (and ZnO-free) production. Nevertheless, truly ABF (or RWA) production, i.e., no antibiotics (or antimicrobials if considering ZnO) provided to the pig through feed, water, or injection during the full course of its life, is challenging. **Key Words:** weaning, gut health, ABF

**187 The nucleoside inosine exerts anti-inflammatory effects and increases cellular energy abundance in porcine intestinal epithelial cells.** Abiola. S Lawal and Kolapo. M Ajuwon*, *Purdue University, West Lafayette, IN, USA*. Intestinal epithelial cells are majorly dependent on the salvage pathway for the synthesis of nucleotides and nucleotides needed for cellular growth and function. This study was conducted to determine the effects of inosine, a major dietary and endogenous nucleoside, on porcine intestinal epithelial cell (IPECJ2) proliferation, maturation, cellular energy content and its anti-inflammatory effects in cells treated with TNFa. Pre-confluent IPECJ2 cells were treated with 0, 250μM, 500μM and 1mM inosine for 72 hours. Cellular proliferation measured with MTT assay showed that inosine dose-dependently ($P < 0.05$) stimulated cell proliferation. Additionally, inosine increased ($P < 0.05$) transepithelial electrical resistance (TEER) and suppressed ($P < 0.05$) FITC-dextran permeability, indicative of the strengthening of barrier integrity of the cells by inosine. Inosine also increased cellular energy content by increasing ($P < 0.05$) the levels of ATP and AMP. Metabolic flux analysis conducted on a Seahorse analyzer indicated that inosine significantly and dose-dependently ($P < 0.05$) increased glycolysis and glycolytic capacity of cells, suggesting that cells increased glycolytic rate in response to inosine, leading to increased cellular energy content. In cells treated with TNFα, inosine (500 μM), significantly attenuated ($P < 0.05$) the disruption of tight junction integrity by preventing the downregulation of TEER and the increase in FITC-dextran permeability caused by TNFα. Additionally, in cells exposed to the mycotoxin deoxynivalenol (DON, 0.5 μM), inosine prevented the fall in TEER and the increase in FITC-dextran passage. Experiments conducted with specific inhibitors of adenosine receptors A1R, A2aR, A2bR and A3R confirmed that activation of the A3R pathway was partly responsible for the observed inosine effects. In conclusion, this study shows the importance of dietary or endogenous nucleosides in the regulation of intestinal development and function. **Key Words:** nucleoside, inosine, epithelial cells

**188 The supplementation of glycerides of lauric acid in sow feed enhances piglet performances at weaning.** A Mellouk[1], V Michel[1], N Vieco[1], O Lemâle[2], T Goossens[3], and J Consuegra*[1], [1]*European Laboratory of Innovation, Science & Expertise (ELISE). Adisseo France S.A.S. R&I in Monogastric Animal Nutrition, Saint Fons, France*, [2]*Adisseo NL B.V., Raamsdonksveer, The Netherlands*, [3]*Adisseo Belgium, Dendermonde, Belgium*. Glycerides of lauric acid supplementation (GLA) in monogastric animals showed a potential immune modulator effect improving cellular and humoral responses to pathogens. This study aims to determine the effects of GLA supplementation in sow feed on the maternal immune transfer and the resilience of piglets to the post-weaning syndrome. Forty-five gestating sows were allocated to two experimental groups, two weeks before farrowing. Their randomization was based on their weight and IgG levels in the blood. The sows received a control or a GLA supplemented diet (2 kg/T) for five weeks: two weeks of gestation and three weeks of lactation. The zootechnical measurements showed comparable average daily feed intake in both sow groups during the gestation (3.8-3.9 kg/d/sow) and the lactation (4.4 kg/d/sow) periods. The sows of both experimental groups gave birth to similar average numbers of born alive piglets: 19 piglets/sow. The piglets from both experimental groups had comparable average BW at birth (1.34 kg and 1.36 kg) and an average of 12 piglets/sow were weaned. Interestingly, piglets from GLA supplemented sows showed 6.6% higher individual weaning weight compared to the control group (4.96 *vs*. 4.65 kg, respectively; $P = 0.019$). Supplementation of GLA in sow feed increased the average daily gain of piglets by 10.2% during lactation (162 vs. 147 g/day, $P = 0.006$). These results show that supplementing sow feed with GLA improves the performance of piglets at weaning. This may be attributed to an improved colostrum and milk quality which contribute the improvement of gut maturation, absorption abilities, mucosal immunity in piglets as well as a better passive immune transfer. To confirm this hypothesis, further analyses are ongoing to assess the maternal immune transfer in the colostrum and milk, the effect of these parameters on the post-weaning syndrome incidence and the piglet health status during this phase.
**Key Words:** Glycerides of lauric acid, Weaned piglets, Passive immune transfer

**189 Influence of Reduced Dietary Crude Protein and Propionic Acid Preservation on Intestinal Health and Growth in Post-Weaned Pigs.** K.R. Connolly*[1], T. Sweeney[2], and J.V. O'Doherty[1], [1]*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland*, [2]*School of Veterinary Medicine, University College Dublin, Dublin, Ireland*. The objective of the study was to determine if organic acid (OA)-preserved grain could alleviate the adverse effects of low crude protein (CP) diets on the growth performance of weaned pigs by improving intestinal health and the coefficient of total tract digestibility (CATTD) of nutrients compared to conventionally dried grain. Grain was either dried conventionally or preserved post-harvest with 4 kg of OA/tonne. Ninety-six 28-day-old pigs were assigned to one of four diets in a 2 × 2 factorial design: (1) dried standard CP diet, (2) OA-preserved standard CP diet, (3) dried low CP diet, and (4) OA-preserved low CP diet. The standard and low CP diets contained 21% and 19% CP for the first 15 days, reducing to 19% and 17.5% CP, respectively, thereafter. On day 8 post-weaning, duodenal, jejunal and ileal tissue was collected for intestinal morphological analysis and QPCR assessment of the expression of multiple inflammatory genes. Ileal and colonic digesta was collected for microbial and volatile fatty acid (VFA) analysis. Pigs and offered feed were weighted weekly for the calculation of average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR). Low CP diets reduced FS ($P < 0.05$) and increased colonic molar butyrate proportions ($P < 0.01$), but elevated duodenal *IL1B* expression compared to standard CP diets ($P < 0.05$). OA-preserved grain enhanced beneficial microbial populations (*Lactobacillus*, *Roseburia*) while decreasing pro-inflammatory cytokines *IL1A* (duodenum), *IL17* (jejunum and ileum) compared to dried grain ($P < 0.05$). While dried grain with low CP diets reduced ADG, colonic VFA concentrations, and nitrogen digestibility, OA-preserved grain with low CP diets maintained these parameters and improved final body weight ($P < 0.05$). Overall, OA-preserved grain mitigated the performance decline linked to low CP diets by promoting gut health and improving nutrient digestibility, offering a promising nutritional strategy for weaned pigs. **Key Words:** weaning, protein, microbiome

**190 Effects of intestinal carnitine transporter expression on carnitine status and fatty acid oxidation in suckling piglets.** T Boston*, F Wang, M Knauer, J Odle, and X Lin, *North Carolina State University*. High neonatal piglet mortality is a major economic and animal welfare issue, partially caused by low carnitine status and inefficient energy generation in neonates. Carnitine is primarily absorbed from the small intestine via active transport, in which organic cation transporters (OCTNs) play an important role. Upregulation of OCTN by activation of PPARs may increase carnitine intestinal absorption. Accordingly, the effects of PPARα (clofibrate) and PPARγ (pioglitazone) agonists on OCTN postnatal expression were evaluated by measuring carnitine status and fatty acid (FA) oxidation. 42 suckling piglets from 6 litters were utilized. Of these, 6 were euthanized within 24 hours after birth (day 1) and the remaining 36 were divided into 3 groups: Control (vehicle: 2% Tween-80), Treatment 1 (vehicle with 75 mg/kg clofibrate), and Treatment 2 (vehicle with 75 mg/kg pioglitazone). Piglets were treated every 2 days via orogastric gavage, and half the pigs were euthanized at day 10 and the other half at day 20. Jejunal relative gene expression showed no detectable increase between treatments ($P > 0.1$), but increased with age for genes related to carnitine transport, *SLC22A4* (7.5 fold), *SLC22A5* (9.2 fold), *SLC6A14* (7.9 fold) and FA metabolism, *PPARα* (2.6 fold), *PPARγ* (4.7 fold), *ACO* (3.9 fold), *FABP1* (2.8 fold; $P < 0.05$). No effects of age or treatment were detected on free, acyl- or total carnitine concentrations ($P > 0.1$), although baseline piglets showed higher total carnitine (0.189 nmol/mg) compared with all other days and treatments (0.067 nmol/mg) ($P < 0.05$). Jejunal β-oxidation rates of oleate (4.85 nmol/mg protein/hour) were similar among treatments ($P > 0.1$); however, there were several interactions of treatment and age ($P < 0.05$). We conclude that intestinal carnitine transporter expression is markedly influenced by piglet age, regardless of PPAR agonists, and is associated with increased rates of FA oxidation. **Key Words:** carnitine, clofibrate, metabolism

**191 Exploring the Combined Benefits of Butyric Acid and Resistant Potato Starch for Gut Health and Metabolism.** K.R. Connolly*[1], T. Sweeney[2], and J.V. O'Doherty[1], [1]*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland*, [2]*School of Veterinary Medicine, University College Dublin, Dublin, Ireland*. This study compared the effects of supplementing exogenous butyrate (BA) as butyric acid versus promoting endogenous BA production via potato starch (PS) supplementation, on the health and growth of weaned pigs. The 96 pigs were sorted into 4 groups and fed for 35 days: (1) control diet (C); (2) C+1.5% BA; (3) C+1% PS; (4) C+1.5% BA+1% PS (BAPS). A faecal score (FS) system of 1-5 was used to monitor diarrhoea (1=very hard, 5=very loose). There was a BA x PS interaction ($P < 0.05$) on daily gain (DG), daily feed intake (FI), final body weight (FBW) and FS. The addition of BA had no effect on DG (510 vs 536, SEM 0.025) or FWB (24.4 vs 25.4, SEM 0.627) compared to the C diet. However, combining BA and PS increased DG (585 vs 518, SEM 0.025) and FBW (27.1 vs 24.9, SEM 0.627) compared to the PS diet. The addition of PS reduced FI compared to the C diet (773 vs 853, SEM 0.027), but combining PS and BA increased FI compared to the BA diet (922 vs 812, SEM 0.027). The addition of BA reduced FS compared to the C diet (2.35 vs 2.50, SEM 0.031), but combining BA and PS increased FS compared to the PS diet (2.27 vs 2.17, SEM 0.031). The inclusion of PS improved ($P = 0.05$) FCR (1.53 vs 1.79, SEM 0.101) compared to non-PS supplemented diets. The addition of PS reduced faecal Proteobacteria (1.84 vs 0.75, SEM 0.359) while increasing faecal *Prevotella* (9.41 vs 3.30, SEM 0.772) and faecal butyrate levels (41.21 vs 26.61, SEM 5.509) compared to non-PS supplemented diets ($P < 0.05$). There was a BA x PS interaction on faecal *Faecalibacterium* ($P < 0.05$); The addition of BA had no effect on *Faecalibacterium* compared to the C diet (8.83 vs 11.57, SEM 1.285), but combining BA and PS increased *Faecalibacterium* compared to the PS diet (16.70 vs 13.38, SEM 1.445). In conclusion, combining BA and PS improved growth performance, faecal scores and gut health. Potato starch supplementation improved faecal scores, feed efficiency, intestinal butyrate levels and modulated the faecal microbiome. **Key Words:** weaning, butyric acid, prebiotic

**192 Evaluating the relationship between weaning weight and early post-weaning feeding behavior.** S. Laird*, L. Sobrevia, L. Blavi, M.A. Ton Nu, A. Koppenol, and S. Tibble, *AB Neo, Fraga, Huesca, Spain*. The continuity of feed consumption after weaning is critical to alleviating the post-weaning growth check and associated disorders. Much of the current understanding of feeding behaviour at weaning has been determined using individually housed pigs, which in itself influences feeding patterns. Using electronic feeding systems, this study set out to evaluate the influence of body weight (BW) on individual feeding behaviour of group-housed pigs immediately post-weaning. At weaning, 100 pigs were selected from a larger pool of pigs based on visual observation to create four BW groups of 25 pigs: 1) Small (**S,** 2.6+0.47 kg); 2) Small-medium (S-M, 5.0+0.60 kg); 3) Medium-heavy (M-H, 6.6+0.72 kg); and 4) Heavy (H, 9.4+0.69 kg). Pigs were ear-tagged with a unique RFID chip and housed within BW group in one of four pens, each equipped with an EFS. To study the development of early post-weaning feeding behaviour, the following characteristics were calculated: latency to first feed; initial feed intake (FI), defined as total consumption in the 24h following the first recorded FI; and the total number of feeder visits in the same 24h period. Initial FI and feeder visit data were analysed by GLM with BW as a fixed effect and pig within pen as a random effect. Latency time was analysed using Kaplan-Meier analysis. Small BW pigs were the quickest to eat after weaning with a mean latency time of 5.3h vs 12.3h for the other BW categories ($P < 0.05$). Moreover, the number of feeder visits significantly decreased with increasing BW ($P < 0.001$). In line with the feeder visit data, an inverse relationship between BW and initial FI ($P < 0.001$) was observed: when standardised for BW, the FI of S pigs was 100%, 175%, and 480% higher than that of the S-M, M-H, and H pigs, respectively. In summary, smaller pigs are quicker to adapt and resume feeding after weaning. Therefore, strategies aimed at improving the early onset of feeding in heavier pigs post-weaning warrant further attention. **Key Words:** Feeding behaviour, weaning, body weight

**193 Thermomechanical and enzyme-facilitated processing of soybean meal enhanced in vitro crude protein digestion kinetics in weaned piglets.** F Njeri*[1], M Anh Ton Nu[2], H Schulze[3], and E. G Kiarie[1], [1]*University of Guelph, Guelph, Ontario, Canada*, [2]*AB Neo, Videbaek, Denmark*, [3]*Livalta, Peterborough, UK*. Feed processing and gastric pH may influence crude protein digestion kinetics in piglets. This study aimed to evaluate the effect of processing and gastric pH on crude protein digestion kinetics in soybean meal (SBM) using *in vitro* model simulating piglets' digestion. Samples (500 mg CP, n=3) of non-processed SBM (SBM, 46.1% crude protein (CP)), thermomechanical and enzyme-facilitated processed soy protein (TES, 53.9% CP) and casein (87.6% CP) were incubated with porcine pepsin at pH 3.5 or 4.5 and at 39°C for 0, 0.5, 1, and 1.5h (gastric phase). After 1.5h gastric phase, all samples were digested with porcine pancreatin and bile extract at pH 6.8 for 0, 0.5, 2, 4, and 6h (small intestinal phase). Data was analyzed with protein source and gastric pH as fixed effects and their interaction. The CP in feed was classified based on their digestion time during the small intestine phase as $CP_{fast}$ (0-0.5h), $CP_{slow}$ (0.5-4h), and $CP_{resistant}$ (>4h). At 1.5h gastric digestion, the interaction ($P < 0.001$) between protein source and pH was such that SBM had similar soluble CP at pH 4.5 but higher soluble CP by 44.3% at pH 3.5 compared to TES. However, after 6h of small intestine digestion, casein and TES had similar soluble CP but higher than SBM by 12.6 and 14.9%, respectively whereas soluble CP released at pH 3.5 was less by 2.2% than at pH 4.5 ($P < 0.05$). Casein and SBM had higher $CP_{fast}$ at gastric pH 3.5 than pH 4.5 ($P < 0.05$) but no gastric pH effect on TES, whereas higher $CP_{slow}$ at pH 3.5 than pH 4.5 was observed in casein ($P < 0.05$) but not in SBM and TES. Casein and TES were similar and had lower ($P < 0.05$) $CP_{resistant}$ than SBM. TES had lower ($P < 0.05$) $CP_{resistant}$ by 48.3% at pH 3.5 and by 40.9% at pH 4.5 than SBM. In conclusion, gastric pH influenced *in vitro* protein digestion kinetics in small intestine at different extent depending on protein source. Processing enhanced crude protein digestion in SBM at either pH 3.5 or 4.5, which may lower protein fermentation in piglets. **Key Words:** Processed soybean meal, *in vitro* digestibility, gastric pH

**194 The supplementation of glycerides of lauric acid in sows' feed enhances piglet performances at weaning.** A Mellouk[1], V Michel[1], N Vieco[1], O Lemâle[3], T Goossens[2], and J Consuegra*[1], [1]*European Laboratory of Innovation, Science & Expertise (ELISE). Adisseo France S.A.S. R&I in Monogastric Animal Nutrition, Saint Fons, France*, [2]*Adisseo Belgium, Dendermonde, Belgium*, [3]*Adisseo NL B.V., Raamsdonksveer, The Netherlands*. Glycerides of lauric acid supplementation (GLA) in monogastric animals showed a potential immune modulator effect improving cellular and humoral responses to pathogens. This study aimed to determine the effects of GLA supplementation in sows' feed on the maternal immune transfer and the resilience of piglets to the post-weaning syndrome. Forty-five gestating sows were allocated to two experimental groups, two weeks before farrowing. Their randomization was based on their weight and IgG levels in the blood. The sows received a control or a GLA supplemented diet (2 kg/T) for five weeks: two weeks of gestation and three weeks of lactation. The zootechnical measurements showed comparable average daily feed intake in both sow groups during the gestation (3.8-3.9 kg/d/sow) and the lactation (4.4 kg/d/sow) periods. The sows of both experimental groups gave birth to similar average numbers of born alive piglets: 19 piglets/sow. The piglets from both experimental groups had comparable average BW at birth (1.34 kg and 1.36 kg) and an average of 12 piglets/sow were weaned. Interestingly, piglets from GLA supplemented sows showed 6.6% higher individual weaning weight compared to the control group (4.96 vs. 4.65 kg, respectively; $P = 0.019$). The supplementation of GLA in sows' feed increased the average daily gain by piglets 10.2% during lactation (162 vs. 147 g/day, $P = 0.006$). These results demonstrate that GLA supplementation in sows' feed strategy improves the piglets' performances. This may be attributed to an improved colostrum and milk quality which contribute to the gut maturation, absorption abilities, mucosal immunity in piglets as well as to a better passive immune transfer. To confirm this hypothesis, further analyses are ongoing to assess the maternal immune transfer in the colostrum and milk, the effect of these parameters on the post-weaning syndrome incidence and the piglet health status during this phase. **Key Words:** Lauric acid glycerides, Weaned piglets, Passive immunity

**195 Impact of sensory flavors and creep feed intake on post-weaning gut barrier function in piglets.** Z.W. Ng'ang'a*[1,2], N. Tous[1], J. Tarradas[1], R. Beltrán-Debón[2], S. López-Vergé[3], J.J. Pastor[3], G. Tedo[3], and D. Torrallardona[1], [1]*IRTA, Animal Nutrition, Constantí, Catalonia, Spain*, [2]*MobioFood Research Group, Universitat Rovira i Virgili, Tarragona, Catalonia, Spain*, [3]*Lucta S.A., Cerdanyola del Vallès, Barcelona, Spain*. This study evaluated the effects of feed sensory flavors and pre-weaning feed intake on gut barrier function in weaned piglets. At d7 of age the litters from 12 sows were assigned to 2 feeding groups: control (T1) and sensory additive (T2), until d14 post-weaning (PW). Piglets were categorized as "eaters" or "non-eaters", before and after weaning using rectal swabs and blue indigo-carmine as feed marker. At weaning, 96 piglets were distributed in 12 pens per group to assess feed intake and performance. Interestingly, T2 had a higher proportion of piglets identified as eaters at d1 PW (69 vs. 46%; $P = 0.02$), had better ADG (156 vs. 129 g/d; $P = 0.04$), tended to improve ADFI and FCR ($P < 0.1$) during the 14d PW period. An additional 48 piglets (12 eaters and 12 non-eaters per feeding group) were selected at weaning to test intestinal permeability at d5 PW: half were tested *in-vivo* with the lactulose, mannitol and ovalbumin tests (oral bolus and plasma concentration after 1h), and the other half were euthanized for *ex-vivo* FITC-dextran intestinal permeability (ap-to-bas) and gut barrier gene expression analyses (microfluidic qPCR) in jejunal and ileal mucosa. The *in-vivo* and *ex-vivo* permeability markers were not affected by treatment, except for an interaction between treatment and eating category for mannitol absorption ($P = 0.02$) where non-eaters benefitted more from the additive. In the ileum, T2 significantly upregulated the gene expression of *OCLN* ($P = 0.01$) and *MUC13* ($P = 0.03$) while an interaction effect of *ZO1* ($P = 0.01$) was observed, suggesting enhanced tight junction integrity and reinforced mucosal defense. Additionally, it upregulated the expression of nutrient transporters *SLC5A1* (glucose, $P = 0.01$) and *SLC39A4* (zinc, $P = 0.02$). These findings highlight the positive effect of a sensory additive to increase feed intake of piglets, support gut barrier function and boost nutrient uptake, thus offering potential strategies to ease their transition at weaning. **Key Words:** Gut barrier, sensory flavors,

weaned piglets

**196 Effects of indigestible dietary protein content on growth performance, immune status, and gut health of nursery pigs.** T. J. Erinle*[1,2], M. J. K. de Oliveira[1], J. K. Htoo[3], S. M. Mendoza[4], and D. A. Columbus[1,2], [1]*Prairie Swine Centre, Inc., Saskatoon, Saskatchewan, Canada*, [2]*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada*, [3]*Evonik Operations GmbH, Rodenbacher Chaussee 4, Hanau-Wolfgang, Germany*, [4]*Evonik Corporation, Kennesaw, Georgia, United States*. The objective of this study was to evaluate the impact of IDP content on performance, immune response, and fecal consistency score (FCS) in newly-weaned pigs. Eighty mixed-sex weaned pigs with an average initial body weight of 8.1 ± 0.31 kg were housed in groups of 5 pigs/pen and randomly assigned to 1 of 2 dietary treatments in a completely randomized design (n = 8 pens/treatment) for 28 d. Corn-soybean diets were formulated to contain similar total CP [22% (phase I) and 20% (phase II)], net energy, and standardized ileal digestibility lysine to meet or exceed nutrient requirements but differ in IDP content [low IDP, 2.6% (LIDP) or high IDP, 4.2% (HIDP)]. On a weekly basis, feed intake and body weight were recorded to estimate average daily gain (ADG), average daily feed intake (ADFI), and Gain:Feed. During the first 7 days and subsequently on a weekly basis, FCS was visually scored. On d 9 and 28 post-weaning, blood samples were collected in the morning from 1 pig/pen without prior fasting. Compared with HIDP, LIDP increased ADG by 7% during phase II and 4% during the overall period ($P < 0.05$). There was no effect of IDP on ADFI ($P > 0.10$). Gain:Feed was lower and higher in LIDP-fed pigs during phases 1 and 2, respectively, compared to HIDP ($P < 0.05$). Pigs fed LIDP diet had improved FCS on d 5 to 7 and overall compared to HIDP ($P < 0.01$). Plasma superoxide dismutase concentration was higher in LIDP-fed pigs compared to HIDP ($P < 0.05$). There was neither IDP, day, nor their interaction effect on serum albumin, *interleukin* (*IL*)-6, *IL*-1β, and plasma alkaline phosphatase ($P < 0.05$). There was an IDP × Day interaction effect on plasma oxidized glutathione (GSSG), which was higher on d 28 in LIDP-fed pigs compared to d 9 ($P < 0.05$); however, was similar to plasma GSSG in pigs fed HIDP on both days. The results indicate that feeding LIDP diet improves growth performance and oxidative status while reducing diarrhea severity in nursery pigs. **Key Words:** indigestible dietary protein, diarrhea severity, acute-phase protein

**197 Impact of therapeutical zinc oxide dietary supplementation on growth performance, fecal score and gene expression of intestinal biomarkers in Postweaning Piglets.** Alberto Torres-Pitarch*[1], Aitor Balfagón[1], Edgar G. Manzanilla[2,3], Juan M. Ortiz Sanjuan[2], Lorcan O'Neilll[2,3], David Solà-Oriol[4], Julia Suppi[4], Encarnación Jimenez-Moreno[1], Richard Faris[1], and Graziano Manotvani[1], [1]*Cargill Animal Nutrition and Health, Schiphol, Netherlands*, [2]*Pig Development Department, The Irish Food and Agriculture Authority, Teagasc, Cork, Ireland*, [3]*School of Veterinary Medicine, University College Dublin, Dublin, Ireland*, [4]*Animal Nutrition and Welfare Service (SNIBA), Department of Animal and Food Science, Autonomous University of Barcelona, Bellaterra, Spain*. The study investigated the effects of therapeutical dietary supplementation of zinc oxide (ZnO) on growth performance, fecal score and gene expression of intestinal biomarkers in post-weaning piglets. A total of 840 piglets (Topigs Norsvin TN70 x Tempo, 26 ± 2.0 days old, 7.4 ± 0.08 kg) were randomly allocated to 2 dietary treatments in 6 pens per treatment. The experiment lasted 14d, the control group (C) received a basal diet and the treatment group (Z) received the basal diet supplemented with 3100 ppm of zinc oxide. Body weight and feed intake were recorded on d0, 7 and 14. Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. On d8 post-weaning, 12 piglets per treatment were humanly euthanized to collect jejunum tissue samples to determine gene expression of a panel of 54 biomarkers related to barrier function, immune response, nutrient transport, enzymes and hormones and stress factors (96.96 Dynamic Array™ IFC, Standard Biotools Inc.). The experimental data was analyzed by ANOVA using SAS® version 9.4. Data are presented as least square means and SEM. Piglets on the Z group had higher ADG than the C group (192 *vs* 162 ± 5.1 g/day, $P > 0.01$), similar ADFI and FCR ($P > 0.10$). The fecal score expressed

as the area under the curve (AUC) was improved in Z piglets (20.0 vs 7.0 ± 0.41 AUC, $P < 0.001$). Out of 54 genes, 10 differed in jejunum expression between treatment groups. Piglets in the Z group showed downregulation of immune-related genes (IL8, CXCL2, IFNGR1, TLR4; $P < 0.05$), as well as reduced expression of stress-related genes (HNMT; $P < 0.05$) and the nutrient transport gene SLC39A4, which is associated with inflammation and zinc uptake dysregulation ($P < 0.05$). Supplementation with therapeutic levels of ZnO appears to enhance growth potential, lower the diarrhea incidence, and support a healthier, functional gut environment by reducing inflammation in the jejunum induced by weaning in piglets. **Key Words:** piglet weaning zinc oxide

**198 Bridging maternal and weaned diets with a continuity in feed plant volatiles has the potential to boost postweaning piglet growth.** Marta Navarro*[1], Gemma Tedo[3], Sergi Lopez[3], Oriol Anglada[3], Viet Hai Tran[1], Sally Taylor[1], Max Muller[1], Amelia Dixon[5], Ryan Clarkson[4], Marion Magnan[4], Katie McDermott[4], Frank R. Dunshea[2,4], and Eugeni Roura[1], [1]*The University of Queensland, Brisbane, Qld, Australia*, [2]*The University of Melbourne, Melbourne, Victoria, Australia*, [3]*Lucta, Sant Cugat, Barcelona, Spain*, [4]*University of Leeds, Leeds, United Kingdom*, [5]*William Thompson, York, United Kingdom*. Post-weaning neophobia and anorexia can lead to low growth rates and compromised health. Plant volatile organic compounds (VOCs) can be transferred from the maternal environment to embryonic tissues, including the amniotic fluid. We hypothesise that continuity in the VOC profiles through gestation, lactation, and weaning diets will positively influence feeding behavior. Sixty sows (Large White × Landrace; parity 3–5) were fed a gestation diet (wheat, barley and soybean meal) enriched with a flavor formulated to replicate and boost the VOC profile from day 80 of gestation. Feed VOCs were determined by HES-SPME/GC-MS. At farrowing, sows were assigned one of two diets: a) an isosensorial (IS) lactation diet, aligned with the gestation diet's VOC profile ([ppm]: hexanal [37.3], methyl amyl ketone [26.3], ethyl hexanoate [17.7], 1-octen-3-ol [5.7], pentanol [4.1], penten-3-ol [2.2], a-pinene [2.1]); or b) a heterosensorial diet (HS) based on corn, rye, legumes and canola meal, with a distinctive VOC profile ([ppm]: propionic acid [86.8], 2,5-dimethyl pyrazine [12.8], isovaleric acid [3.4], heptanal [0.6], nonanol [0.5], ß-pinene [0.2]). A 2 × 2 factorial design assessed lactation (L-IS or L-HS) and weaning (W-IS or W-HS) diets. IS and HS diets were isoaminoacidic and isoenergetic. At weaning, 320 piglets were divided into four experimental groups (five animals/pen, balanced by weight, gender and litter). Average daily gain (kg/d) was higher in W-IS-fed pigs [0.151 (L-IS), 0.168 (L-HS)] than in W-HS-fed pigs [0.126 (L-IS), 0.119 (L-HS)] ($P < 0.0001$). Feed conversion was lower in the W-IS [1.489 (L-IS), 1.362 (L-HS)] than in the W-HS [1.754 (L-IS), 1.741 (L-HS)] ($P < 0.0001$). The lactation diet and its interaction with the weaner diet had no significant effects. Maintaining consistent VOC profiles across sow and piglet diets may reduce dietary neophobia and improve post-weaning growth and feed efficiency. **Key Words:** imprinting, post-weaning, VOCs

**199 Effects of indigestible dietary protein content on growth performance, immune status, and gut health of nursery pigs challenged with enterotoxigenic *Escherichia coli* F4 or *Salmonella* Typhimurium.** T. J. Erinle*[1,2], M. J. K. de Oliveira[1], K. S. Ng[2], R. D. Kim[2], J. C. Panisson[1], J. K. Htoo[3], S. M. Mendoza[4], J. L. Thomassin[5], and D. A. Columbus[1,2], [1]*Prairie Swine Centre, Inc.,, Saskatoon, Saskatchewan, Canada*, [2]*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada*, [3]*Evonik Operations GmbH, Rodenbacher Chaussee 4, Hanau-Wolfgang, Germany*, [4]*Evonik Corporation, Kennesaw, Georgia, United States*, [5]*Department of Biochemistry, Microbiology and Immunology, University of Saskatchewan, Saskatoon, Saskatchewan, Canada*. The objective of this study was to examine the effect of indigestible dietary protein (IDP) content on growth performance, immune status, and fecal consistency score (FCS) of nursery pigs challenged with enterotoxigenic *E. coli* K88$^+$ F4 (ETEC) or *Salmonella* Typhimurium (ST). Thirty-two mixed-sex nursery pigs with an average initial body weight of 7.26 ± 0.40 kg were individually housed and randomly assigned to 1 of 2 dietary treatments in a completely randomized design (n = 8 pigs/treatment) for 14 days (d). Diets were formulated to

contain similar crude protein (CP) content (21%), but differed IDP content [low IDP, 2.6% (LIDP) or high IDP, 4.2% (HIDP)]. After a 7-d pre-inoculation period, pigs were orally inoculated with either $1.36 \times 10^6$ CFU·mL$^{-1}$ ETEC or $1.14 \times 10^{10}$ CFU·mL$^{-1}$ ST (n = 16 pigs/challenge). Growth performance, rectal temperature, FCS, and blood inflammatory biomarkers were measured pre- and post-inoculation. There was no dietary effect on any measures pre-inoculation ($P > 0.05$). Inoculation with ETEC F4 or ST increased rectal temperature, FCS, and blood inflammatory cytokines, acute-phase proteins, and redox biomarkers ($P < 0.05$). Unlike in ETEC-pigs ($P > 0.05$), LIDP increased average daily feed intake and average daily gain ($P < 0.05$) and tended to increase gain:feed ($P < 0.10$) compared to HIDP-fed pigs inoculated with ST. Likewise, unlike in ETEC-pigs, LIDP diet reduced FCS compared with HIDP-fed pigs ($P < 0.05$) under ST challenge. Regardless of enteric pathogens, HIDP increased serum *interleukin*-6 and plasma haptoglobin compared to LIDP ($P < 0.05$). In ST-pigs, HIDP further increased serum albumin, tumor necrosis factor-alpha, and diamine oxidase and pathogen translocation to mesenteric lymph node, while reducing plasma reduced glutathione (GSH) and GSH:oxidized glutathione ($P < 0.05$). The results indicate that increasing dietary IDP content resulted in greater negative outcomes on performance, fecal score, and inflammation in ST-pigs compared with ETEC-pigs. **Key Words:** post-weaning, enteric pathogenic infection, diarrhea severity

**200 Comparing the effects of a high dose of acidifiers to those of a double encapsulation of bioactives on growth and gut microbiota of post-weaning piglets.** S. Ladirat*[1], V. Bernad[2], M. Mallen[2], and S. Nadal[2], [1]*NUQO S.A.S, Annecy, France*, [2]*Test & Trials, Monzón, Spain*. Organic acids and plant extracts are widely used to support post-weaning piglet growth, but their composition, form, and dosage can yield varying outcomes. This study compared the effects of a double-encapsulated solution containing organic acids (core) and plant extracts with algae meal (outer layer) against a high dose of non-encapsulated acidifiers on piglet performance and health over 42 days post-weaning. A total of 231 piglets (50% Hybird DanBred x 50% German Pietrain) weaned at 24 days were housed in 33 pens of 7 males, grouped by initial body weight. Treatments included: 1. basal diet (NC), 2. basal diet with double-encapsulated solution at 800 g/t (SF), and 3. basal diet with acidifiers at 8 kg/t (days 1 to 14) and 4 kg/t (days 14 to 42) (AB). Each treatment had 11 replicates. On days 14 and 42 post-weaning, zootechnical performance was recorded per pen while microbiota composition and short-chain fatty acids (SCFA) were assessed using fecal samples from 11 piglets per treatment. Data were analysed via GLM (productive parameters) and ANOVA (log-transformed microbiota and SCFA). In the grower phase (days 14 to 42) and overall (days 1 to 42), piglets fed AB showed lower feed intake and body weight compared to NC ($P < 0.05$), indicating palatability issues. Piglets fed SF exhibited improved FCR over NC ($P < 0.05$). On day 14, SF reduced total E. coli relative abundance while AB increased it compared to NC ($P < 0.05$). AB also increased the relative abundance of *Enterotoxigenic E. Coli*, decreased that of *Lactobacillus* and showed reduced lactic acid levels compared to NC ($P < 0.05$). On day 42, SF increased *Lachnospiraceae* relative abundance, correlating with higher butyrate levels ($P < 0.05$). These findings show that, unlike a high dose of non-encapsulated acidifiers, the double encapsulation promoted butyrate-producing bacteria, suppressed pathogens, and enhanced piglet feed efficiency, indicating improved gut preparation for further performance till slaughter. **Key Words:** post-weaning, piglet performance, microbiota composition

**201 The degree of intrauterine growth restriction influences intestinal gene expression and histomorphology in newborn piglets.** P. Salgado-López*[1], C. Soldevila[2], J. Gasa[1], and D. Solà-Oriol[1], [1]*Animal Nutrition and Welfare Service (SNIBA), Department of Animal and Food Science, Autonomous University of Barcelona, Bellaterra 08193, Spain*, [2]*Vall Companys Group, 25191 Lleida, Spain*. This study tested the hypothesis that the severity of intrauterine growth restriction (IUGR) affects intestinal development and function, potentially contributing to reduced growth performance and increased morbidity. Litter and piglet characteristics were monitored at farrowing from 102 hyperprolific sows (Landrace x Yorkshire) and 86 non-hyperprolific sows (Landrace x Large White) across two herds. Just after birth, 48 piglets per herd were selected

for jejunum and ileum sampling. Birth weight (BIW) categories were defined as severe IUGR (sIUGR, ≤ 750 g, n = 12), low-birthweight (LBW, > 750 and ≤ 1150 g, n = 24), and normal-birthweight (N, > 1150 g, n = 12). LBW piglets were further classified into restricted LBW (R-LBW) and non-restricted LBW (NR-LBW) based on the mean value of an index, $\alpha * Z_{BMI} + \beta * Z_{DIFF}$, used as the cutoff point, where $Z_{BMI}$ was the standardized body mass index and $Z_{DIFF}$ was the standardized difference between the piglet's BIW and the average litter weight at birth. The expression of 43 genes in jejunal tissue was analyzed by RT-qPCR using a microfluidic dynamic array. Villus height (VH), crypt depth (CD), and the villus height-to-crypt depth ratio (VH:CD) were measured in each ileum sample. At birth, VH was higher ($P < 0.001$) in N piglets than in R-LBW and sIUGR piglets. Similarly, CD was greater ($P < 0.05$) in N piglets compared to sIUGR piglets. VH:CD ratios were higher ($P < 0.05$) in N piglets than in sIUGR piglets from hypeprolific sows. Piglets with sIUGR exhibited up-regulation of genes related to immune response (*i.e.*, TLR4, TGFB1, NF-Kb) and stress (*i.e.*, CRHR1, HSD11B1) compared to their heavier littermates ($P < 0.05$). Similar gene expression patterns were observed between NR-LBW and N piglets, as well as between R-LBW and IUGR piglets. In conclusion, increasing IUGR severity is associated with impaired gut development and distinct intestinal gene expression patterns in affected piglets compared to their heavier littermates at birth. **Key Words:** gene expression, gut development, intrauterine growth restriction

**202 Born to battle: analyzing sex differences in early survival of intra-uterine growth restricted and normal birth weight piglets.** M. Loyens*, L. Van Bockstal, S. Prims, S. Van Cruchten, and C. Van Ginneken, *Comparative Perinatal Development, Department of Veterinary Sciences, Faculty of Biomedical, Pharmaceutical and Veterinary Sciences, University of Antwerp, Wilrijk, Antwerpen, Belgium*. Hyperprolific sows have a higher prevalence of piglets born with intra-uterine growth restriction (IUGR). These piglets commonly face increased pre-weaning mortality. Male piglets appear more vulnerable in stressful situations, such as IUGR, potentially contributing to their higher pre-weaning mortality rates. This study aimed to examine the influence of sex on neonatal survival and behavior in normal birth weight (NBW) and IUGR piglets. Early identification of high-risk piglets is crucial for targeting interventions to reduce pre-weaning mortality. At a commercial pig farm, 161 sow parturitions were monitored. From all born piglets, we noted their sex, birth hour, birth order, and litter size. Piglets were categorized based on their birth weight and phenotypic characteristics. NBW piglets had a birth weight within the mean birth weight ± 1 standard deviation (SD) and no IUGR characteristics, while IUGR piglets had a birth weight below the mean birth weight – 1 SD in combination with an IUGR phenotype (steep, dolphin-like forehead, bulging eyes, and mouth wrinkles). Per litter, each IUGR piglet was sex-matched with an NBW piglet. From 240 NBW (male: 128; female: 112), and 248 IUGR piglets (male: 130; female: 118) rectal temperature was recorded at 0h, 1h, 3h, and 24h after birth. The selected piglets were observed from birth until the time it took them to reach the udder (U), a functional teat (FT), and start suckling (S). Linear mixed modelling revealed that IUGR piglets performed significantly worse than NBW piglets in thermoregulation (T), latency measurements, weight gain (WG), and colostrum intake (CI) (all $P < 0.0001$). No sex differences were seen in this dataset (T: $P = 0.350$; U: $P = 0.332$; FT: $P = 0.056$; S: $P = 0.824$; WG: $P = 0.836$; CI: $P = 0.609$). To conclude, while IUGR piglets exhibit poorer performance and require targeted interventions, this study highlights the need to explore whether these interventions alone are sufficient. **Key Words:** IUGR, sex differences, neonatal survival

**203 Coarsely ground oat hulls affect gastrointestinal tract development in weanling pigs.** TG Hulshof, HMJ van Hees*, and MO Wellington, *Trouw Nutrition R&D, Boxmeer, The Netherlands*. The objective was to study the effect of feeding diets with high insoluble dietary fiber by including 15% oat hulls (OH), either ground fine or coarse, on gastrointestinal tract development in weanling pigs. A total of 72 pigs (26 ± 0.6 days; 7.0 ± 1.2 kg BW) were housed in 3 pens equipped with 3 electronic feeding stations each containing one of three experimental diets. Pigs had access to only one of three experimental diets. Diets consisted of finely ground highly digestible ingredients (CON; 10.4 MJ NE, 12.5 g/kg SID

Lys), or 15% finely ground (OHf) or 15% coarsely ground OH (OHc) replacing corn starch (9.4 MJ, 12.3 g/kg SID Lys). Growth performance was determined and 12 randomly selected pigs/diet were euthanized on d13 post-wean for intestinal measurements. Empty organs and their contents were weighed and expressed in g/kg BW at dissection, stomach content was analyzed for pH and DM in the proximal and distal area, and apparent ileal digestibility of crude protein and fat were determined. Data were analyzed using MIXED models with treatment as fixed effect and pen as random effect. No differences were observed for growth performance ($P > 0.05$). Empty stomach weight was highest for OHc, followed by OHf, and CON (9.66, 8.78, and 7.32 g/kg BW, respectively; $P < 0.01$). Weights of gastric content ($P = 0.03$), cecum content ($P = 0.03$), empty colon ($P = 0.03$), and colon content ($P = 0.02$) were higher for OHf and OHc compared to CON. Pigs fed the OHc diet had a higher pH gradient (0.93; $P < 0.01$) and smaller DM gradient (-2.3%-point; $P = 0.03$) between the proximal and distal area of the stomach compared to CON (-0.12 and -9.4%-point, respectively) and OHf (0.23 and -4.3%-point, respectively) caused by higher pH and DM in the proximal stomach. Apparent ileal digestibility of crude protein and fat were not different ($P > 0.05$) between treatments. In conclusion, including OH, mainly coarsely ground, affected stomach development without affecting nutrient digestibility and growth. **Key Words:** Stomach, oat hulls, weanling pig

**204 The power of dairy: the effect of milk protein and different lactose levels in creep feed on the pre- and post-weaning performance of piglets.** I.M. Van As*, P.T. van 't Veld, and L.C.M. van Enckevort, *Denkavit Nederland BV, Voorthuizen, The Netherlands*. Over two trials, it was hypothesized that providing creep feed (CF) with milk protein (MP) and/or lactose would stimulate pre-weaning intake and thereby improve growth performance pre- and post-weaning. CF diets were provided from day 3 after birth until weaning at 27 days in both trials. In the 1st trial, 60 litters were divided into 2 groups (32 vs. 28) in 2 replicates. Litters received either CF with 2% MP (A) or CF without MP (B), whereas lactose levels were equal. In the 2nd replicate 46 piglets (24 vs. 22) were selected at 21 days to be subsequently monitored in a digestibility study. Piglets received the same CF as pre-weaning for 4 days and thereafter all piglets received a similar weaner diet. Feed- and water intake, and fecal- and urine samples were collected from day 8-12 post-weaning. In the 2nd trial 62 litters were allocated into 3 groups (24 vs. 20 vs. 18). Groups received either 3 (A), 9 (B) or 15% (C) lactose, whereas MP levels were equal in CF diets. Data was analysed in a ANCOVA model with either birthweight (pre-weaning) or weaning weight (post-weaning) added as a covariate. In the 1st trial a trend was found for higher CF intake for group A up to day 20/21 (177 vs. 129 g/piglet; $P = 0.05$), but this did not result in higher growth. In the post-weaning period, the N absorption, N retention, fat absorption and DM absorption were similar. In the 2nd trial, piglets fed CF with 9% lactose had higher CF intake than piglets fed CF with 3% and 15% lactose between 20-27 days (461 vs. 587 vs. 551 g/piglet; $P = 0.02$). However, no difference in weight gain was found. This indicates that lactose inclusion impacts on CF intake of piglets, suggesting a palatability effect. Inclusion of MP or increased inclusion of lactose, up to an optimum, stimulated piglets towards early consumption of CF, but did not result in higher growth. Improvement of pre-weaning weight gain as a result of higher CF intake may depend on combining optimal lactose and MP levels. **Key Words:** Dairy, Creep feed, Suckling- and weaned piglets

**205 Slow- compared to fast-growing piglets have reduced feed intake and poorer feed conversion in the first 14 days after weaning.** P. Bogere*[1], M. Navarro[1], J. Pluske[2], and E. Roura[1], [1]*Centre for Animal Science, Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Brisbane, Queensland, Australia*, [2]*Faculty of Science, The University of Melbourne, Melbourne, Victoria, Australia*. High growth variation in piglets causes significant economic challenges to the industry. Early feed intake following weaning plays a crucial role in shaping gut architecture and influencing production. This study investigated the growth performance and average daily feed intake (ADFI) of weaned piglets over the first 2 weeks after weaning. It was hypothesised that lighter piglets at weaning would have lower ADFI, slower average daily gain (ADG), and greater growth variation compared to heavier piglets. Ninety-eight intact male piglets from 13 sows of parity

2–5 were weaned at 20 days of age. Twelve healthy piglets were selected from the top 15% (average: 6.9 ± 0.5kg) and 12 from the bottom 15% (average: 3.2 ± 0.6kg) of the weight distribution. The 24 piglets were individually penned with enrichment and fed a standard weaner diet on an *ad libitum* basis (ME: 14.8 MJ/kg, SID Lys: 13.2 g/kg). The daily feed intake and BW (measured every two days) were recorded. Data were tested for normality, and Welch Two sample t-tests as well as Pearson's correlation were performed in R studio. By day 14, fast-growing piglets (FG) had a higher ADG compared to slow growing (SG) piglets ($P < 0.0001$). The variability in ADG was also greater in FG (SD = 43.72 g/day) compared to SG piglets (SD = 20.98 g/day). However, the coefficient of variation (CV) was similar in both groups, 22% and 23% ($p > 0.05$) respectively. Over the 14 days, FG piglets had higher ADFI ($P < 0.001$) and lower feed conversion ratio (FCR) ($p = 0.001$) than SG piglets. In both groups, ADG increased linearly ($P < 0.001$) with higher ADFI. However, ADFI was negatively correlated with FCR ($P < 0.05$), and FCR was negatively correlated with ADG ($P < 0.001$). Weaning weight was positively correlated with both ADFI and ADG ($P < 0.001$). In conclusion, slow compared to fast-growing piglets exhibited lower feed intake, poorer feed conversion, and lower ADG. However, variability in growth was proportional to the mean ADG in both groups. **Key Words:** Feed intake, Growth, Variation

**206 The ratio between SID Thr and total dietary fiber in the weanling pig diet impacts intestinal morphology and mucin secretion.** M.O. Wellington*, T.G. Hulshof, and H.M.J. van Hees, *Swine Research Centre, Trouw Nutrition R&D, Veerstraat 38, 5831 JN, Boxmeer, Netherlands*. Increasing mucus secretion through feeding of high total dietary fiber (TDF) with adequate dietary threonine (Thr) may be used to enhance intestinal barrier function. The objective of the study was to determine the impact of increasing TDF while compensating for SID Thr/Lys on weanling pig intestinal morphology and fecal mucin content. Weaned piglets (6.4 ± 0.3 kg) were housed (3 pigs/pen) and assigned to 1 of 5 dietary treatments (n = 18 pens/treatment) for a 38-d period. Diets consisted of different TDF g/kg, and SID Thr/Lys ratios; (T1) TDF145/Thr0.63, (T2) TDF165/Thr0.63, (T3) TDF165/Thr0.75, (T4) TDF175/Thr0.75 and (T5) TDF175/Thr0.81. A common commercial diet was fed during the first 10 d postweaning, and the experimental diets were fed for 28 d. On d 38, pigs (10 pigs/treatment) were euthanized for ileal and jejunal tissue and fecal samples, for villi height and crypt depth, goblet cell numbers and size, and fecal mucin analyses. Data were analyzed as a one-way ANOVA with treatment as a fixed effect. Ileal goblet cell (**GC**) size was higher in T3 and T4 ($P < 0.05$), while GC numbers were lower for T3 and T4 compared to the other treatments ($P < 0.05$). Jejunal GC numbers were not different ($P > 0.05$), but GC size was higher in T1 compared to T2 and T4, but similar to T3 and T5. Ileal villi height and crypt depth were not different ($P > 0.05$). In the jejunum, crypt depth was higher in T1 compared to T2 and T4, but not different from T3 and T5. A higher fecal mucin was observed in T3 compared to T2 and T5 ($P < 0.05$), whereas, T3 was not different from T1 and T4 ($P > 0.05$). Thus, a higher SID Thr/Lys was required to support higher mucin secretion, with the same TDF level (T3 vs T2; $P < 0.05$). A higher TDF (T5) did not enhance mucin secretion, compared to T3. In summary, the benefit of increasing TDF to stimulate mucin secretion is supported at a higher dietary SID Thr level, hence, a ratio between TDF/SID Thr may help optimize the effect of high dietary fiber on mucin secretion. **Key Words:** dietary fiber, threonine, mucin

**207 Effects of piglet weaning weight on growth performance, hematological parameters, plasma antioxidant status and gut permeability in early nursery period.** C. H. Kwon*, E. Safaie, J. Torres, and Y. D. Jang, *University of Georgia, Athens, GA, USA*. This study was conducted to investigate the effect of piglet weaning weight (WW) on growth performance, hematological parameters, antioxidant status, and gut permeability in the early nursery period. At weaning, a total of 48 pigs (24 barrows and 24 gilts) weaned at 20.7 ± 0.74 d of age were allotted to 2 WW categories in 3 replicate pens per treatment with 8 pigs/pen for 14 d postweaning period. Treatments were: 1) HWW: high WW over 6.0 kg (average weight: 6.79 ± 0.53 kg) and 2) LWW: low WW less than 5.0 kg (average weight: 4.43 ± 0.56 kg). Growth performance, hematological parameters,

antioxidant status, and diamine oxidase levels were measured and analyzed by t-test. In d 0-14 postweaning, the HWW pigs had a greater growth than the LWW pigs ($P < 0.05$; 0.161 and 0.126 kg/d, respectively), resulting in greater body weight on d 14 postweaning ($P < 0.05$; 9.04 and 6.20 kg, respectively). On d 7 postweaning, the HWW pigs had greater mean platelet volume ($P < 0.05$; 10.47 and 9.71 fl, respectively) and lymphocyte percentage ($P < 0.05$; 54.08 and 44.46%, respectively) but lower monocyte count ($P < 0.05$; 0.86 and 1.21 $10^3$/µL, respectively) than the LWW pigs, while no differences were observed in the other hematological parameters. On d 14 postweaning, the HWW pigs tended to have greater plasma superoxide dismutase activity than the LWW pigs ($P = 0.05$; 5.07 and 3.97 U/mL, respectively), with no difference in plasma malondialdehyde levels. Plasma superoxide dismutase activity on d 14 postweaning had a positive correlation with pig body weight from weaning to d 14 postweaning ($P < 0.05$; $R^2 = 0.47$ to 0.68). The HWW pigs had lower plasma diamine oxidase ($P < 0.05$; 27.42 and 37.15 ng/mL, respectively) and d-lactate levels ($P = 0.06$, tendency; 0.74 and 1.46 mM, respectively) than the LWW pigs on d 14 postweaning, indicating that the LWW pigs had greater gut permeability. In conclusion, the HWW pigs had greater growth rate and antioxidant capacity but lower gut permeability than the LWW pigs in the early nursery period. **Key Words:** antioxidant status, gut permeability, pig weaning weight

**208 How does protein level in the maternal diet affect colonic metabolite profile and microbiota in the offspring?** Y. Li[1], K. Kroeske[2,3], M. Schroyen[3], S. Millet[2], C. Van Poucke[2], N. Everaert*[1], and M. Heyndrickx[2], [1]*KU Leuven, Heverlee, Belgium*, [2]*ILVO, Melle, Belgium*, [3]*Liège University, Gembloux, Belgium.* Reducing crude protein (CP) with amino acid addition has shown benefits for sustainable swine production. This study investigated how maternal and piglet CP levels interact to affect the offspring's gut metabolites and microbiota composition. Thirty-five sows were fed a high (17%) or a low (12%) CP diet from day 80 of gestation until farrowing. From weaning at 3.5 to 9 weeks of age (WoA), half of the piglet pens from both groups were given a high CP (17%) diet, and the other half were given a low CP diet (12%), resulting in a 2 × 2 factorial design: HH: Both sows and piglets fed a high CP diet. HL: Sows fed a high CP diet, piglets a low CP diet.; LH: Sows fed a low CP diet, piglets a high CP diet; LL: Both sows and piglets fed a low CP diet. At 3.5, 4.5, and 9 WoA, the colonic digesta of piglets were collected to analyze metabolites and microbiota composition. The low CP in maternal diet significantly increased the colonic concentrations of isovalerate and biogenic amines (BA; including cadaverine, histamine, tryptamine, and tyramine; $P_{sow}<0.05$), and reduced the relative abundances of Bacteroidetes ($P_{sow} = 0.064$), Euryarchaeota ($P_{sow} = 0.072$), and *Bacteroides* ($P_{sow} =0.046$) at weaning. At 4.5 WoA, in the LH group isobutyrate, putrescine, tyramine, and total BA, were the highest among the four groups, and all significantly higher than in the HH group ($P_{interaction} < 0.05$), suggesting that the maternal CP level indeed modulated the protein fermentation of the progeny. At 9 WoA, the LH piglets had significantly higher total BA levels ($P_{interaction} =0,026$) than the piglets in the HL and LL groups suggesting more protein fermentation occurred. At 9 WoA, the HH and HL piglets had significantly different colonic microbiota structures ($P_{interaction} < 0.05$) due to their different diets. In conclusion, maternal dietary protein levels influenced the offspring's colonic metabolite profile, with interactions between maternal and offspring protein levels shaping this profile. **Key Words:** maternal programming, microbial metabolites, microbiota

**209 From preterm piglets to preterm newborns: Investigating the role of postconceptional and postnatal age on hepatic CYP3A and UGT enzyme activity.** L. Buyssens*[1], A. Valenzuela[1], S. Prims[1], M. Ayuso[1], T. Thymann[2], C. Van Ginneken[1], and S. Van Cruchten[1], [1]*Comparative Perinatal Development, Department of Veterinary Sciences, Faculty of Pharmaceutical, Biomedical and Veterinary Sciences, University of Antwerp, Wilrijk, Belgium*, [2]*Comparative Pediatrics and Nutrition, Department of Veterinary and Animal Sciences, University of Copenhagen, Frederiksberg, Denmark.* Recently, the understanding of drug metabolism in the human pediatric population has significantly improved, but data in preterm neonates remain scarce. Enhancing our knowledge of administration, distribution, metabolism and excretion properties in this

vulnerable group is critical to prevent suboptimal dosing. In addition, the impact of perinatal age (expressed as postconceptional (PCA) and postnatal (PNA) age) on these parameters is poorly understood. The juvenile pig serves as an effective model for hepatic drug metabolism in human neonates and infants. However, the impact of prematurity on the onset of hepatic phase I and phase II enzyme activity has yet to be explored. The aim of this study was to assess the ontogeny of cytochrome P450 3A (CYP3A) and uridine 5′-diphospho-glucuronosyltransferase (UGT) enzyme activity in the liver of preterm (gestational day (GD) 105–107) and term-born (GD 115–117) domestic piglets. In addition, the potential effect of PCA or PNA on the onset of enzyme activity was assessed. The following age groups were included: preterm postnatal day (PND) 0 (n = 10), PND5 (n=10), PND11 (n = 8), PND26 (n = 10) and term PND0 (n = 10), PND5 (n = 10), PND11 (n = 8), PND19 (n = 18) and PND26 (n = 10). Liver microsomes were extracted, and a luminescent and fluorescent assay assessed CYP3A and UGT-specific enzyme activity, respectively. CYP3A activity was significantly affected by age ($P$ = 0.0402) and group ($P$ = 0.0021). However, preterm CYP3A activity was only detectable at PND26, whereas term CYP3A activity showed a gradual postnatal increase from PND11 onwards. UGT activity gradually increased between PND0 and PND26 in preterm and term-born piglets, albeit, being systematically lower in the preterm group ($P$ < 0.0001). Thus, PCA is suggested as the main driver affecting porcine CYP3A and UGT enzyme ontogeny. In conclusion, decreased hepatic CYP3A and UGT enzyme activity was detected in preterm-born piglets which may affect their overall biotransformation capacity. **Key Words:** preterm, pig, drug metabolism

**210 Effects of tributyrin supplementation alone or combined with a *Bacillus subtilis* probiotic on performance and gut health of nursery pigs.** B Jayaraman[1], L.V Kinh[2], N.V.T.H Loan[2], L Bauer[3], and J.K. Htoo*[3], [1]*Evonik Methionine (SEA) Pte. Ltd., Singapore, Singapore, Singapore*, [2]*Faculty of Veterinary and Animal Sciences, HUTECH University, Ho Chi Minh city, Vietnam*, [3]*Evonik Operations GmbH, Hanau-Wolfgang, Germany*. A study was conducted to evaluate the effect of supplementing tributyrin (TB) alone or combined with a *Bacillus subtilis* probiotic on performance and gut health status in weaned pigs. Eight hundred weaned piglets (Large white Yorkshire x Landrace; 5.95 kg initial body weight (BW)) were randomly assigned to 4 dietary treatments: (1) a nutrient-adequate basal diet based on corn, soybean meal and whey as negative control (NC; Pre-starter diet: 1.42% SID Lys and 2,488 kcal/kg NE, Starter diet: 1.32% SID Lys and 2,478 kcal/kg NE), (2) NC + antibiotic growth promoter (AGP: colistin & doxycycline at 0.06%) as positive control (PC)), (3) NC + TB (0.15% in Pre-starter and 0.12% in Starter feed (TB), and 4) NC + TB (0.10% in Pre-starter and 0.08% in Starter) and *Bacillus subtilis* probiotics (0.05% in both phases) (TB+PB). Piglets had *ad libitum* access to mash feed and drinking water during the experimental period of 42 days (d). The pre-starter phase was from d 0 to 21 and starter phase from d 22 to 42. The PROC GLM procedures of SAS were used to analyze all data in a randomized block design, and pen was the experimental unit. Results indicated that pigs fed PC had improved ($P$ < 0.05) the final BW and FCR throughout the study, however, dietary supplementation with TB and TB+PB increased ($P$ < 0.05) the final BW compared to NC (25.2 kg, 25.7 kg versus 23.9 kg). FCR of pigs fed TB and TB+PB was better ($P$ < 0.05) than that of NC (1.48, 1.44 versus 1.56). The incidence of diarrhea and fecal score were improved ($P$ < 0.05) in pigs fed PC, TB and TB+PB compared with NC. Overall, FCR and fecal score of pigs were improved by combined supplementation with a tributyrin and probiotic compared with adding tributyrin alone. Based on these results, supplementation with either TB or TB+PB could be beneficial to reduce diarrhea rate and improve performance in nursery pigs fed AGP-free diets. **Key Words:** tributyrin, probiotic, weaned pigs

**211** ***Evaluation of different feeding strategies for underweight weaning piglets: Effect of feed program, feed form and diet composition.*** L Blavi, L Sobrevia, S Laird, S Tibble, and A Koppenol*, *AB Neo, Fraga, Huesca, Spain*. Highly prolific hybrid lines result in more underweight piglets weaned, which require specific nutritional adaptations. Therefore, the trial aimed to evaluate different nutritional strategies during the first week post weaning on piglet performance and health. Different feed forms (mash vs pellet), feed program (4-phase vs 3 phase) and diet composition (standard vs highly digestible ingredients) were evaluated in phase-1 diet. A total of 1200 underweight weaned piglets (24d of age, 3.93±0.71kg) were randomly distributed over 48 pens, 25 pigs each. Body weight and feed intake was recorded, to calculate the average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR). Survivability, morbidity and medication usage were recorded daily. Each of the feeding strategies were considered as fixed factors in the model. No differences were observed in performance between the feed programs. However, the 4-phase feed program resulted in a reduction of medicated ($P$ = 0.04) and removed animals ($P$ = 0.01). ADFI was higher for the mash compared to pellet diet (53.5 g/d more, $P$ < 0.01) during phase-1, resulting in a higher overall FCR (0.051 points, $P$ = 0.04). Medication use was lower in piglets fed pellet diets ($P$ = 0.05). ADFI was higher in the highly digestible diet compared to the standard diet (51.55 g/d more, $P$ < 0.01), resulting in a higher ADG (20.64 g/d, $P$ = 0.02), but also in a higher FCR (0.05 points, $P$ < 0.01) during first week post weaning, higher overall survivability (3.67%, $P$ < 0.01) and lower medication usage (13% less, $P$ < 0.01). The different feeding strategies affecting postweaning performance and health highlighted the importance of specific nutritional adaptations for underweight piglets from hyper prolific sows in the critical postweaning period. **Key Words:** underweight piglets, feeding strategies, weaning performance

**212** **Evaluation of the fecal inflammatory biomarkers' calprotectin and lipocalin evolution through the nursery period in piglets.** J. Suppi*[1], E. Llauradó-Calero[1], C. Soldevila[2], A. Pelegrí-Pineda[3], Y. Saco[3], A. Bassols[3], and D. Solà-Oriol[1], [1]*Animal Nutrition and Welfare Service (SNIBA), Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain*, [2]*Vall Companys Group, 25191 Lleida, Spain*, [3]*Veterinary Clinical Biochemistry Service (SBCV), Department of Biochemistry and Molecular Biology, School of Veterinary, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain*. The nursery period involves significant challenges for piglets due to weaning stress and the decline of passive immunity, often resulting in uncontrolled intestinal inflammation. However, non-invasive methods to assess gut inflammation in piglets are limited. This study aimed to evaluate the efficacy of the fecal inflammatory biomarkers (fBM) calprotectin (fCal) and lipocalin (LCN) in assessing these inflammatory challenges. To model extremes of inflammation, piglets were divided into two dietary groups: a control group (Con) and CON with added zinc oxide (ZnO; 1000 mg/kg of feed). A total of 18 piglets ([L x YS] x Pt), weaned at 23 ± 2 days of age (5.21 ± 1.08 kg) were used for fecal sampling. Half of the piglets were fed a diet containing ZnO for the first 14 days post-weaning (PW), while the other half received a diet without. After this period, all piglets were given the same diet without ZnO. Piglets were weighed and fecal samples were collected on days 3, 8, 14, 21, and 28 PW. The fCal levels were measured using an immunoturbidimetric method, and LCN levels via ELISA. Body weights (BW) were analyzed using a linear model, while fBM levels were assessed longitudinally using a linear mixed-effects model (R software). During the first two weeks PW, piglets receiving ZnO had higher BW, achieving 700 g more by day 8 PW (P = 0.022). After this period, no significant differences in BW were observed ($P$ > 0.05), although ZnO piglets weighed 200 g more on both days 21 and 28 PW. Both fBM showed a quadratic trend ($P$ < 0.055), starting and ending with higher values and with the lowest point by day 14 PW. The ZnO group exhibited lower fCal and LCN values initially but surpassed the control group by the end of the nursery ($P$ < 0.022). The evolution of the fBM aligns with the hypothesis regarding the nursery period, with higher initial values reflecting weaning stress and a second peak after day 14 PW, likely linked to the feed change and immune response maturation. **Key Words:** gut health, non-invasive markers, post-weaning diarrhea

**213 Analysis of specific fecal biomarkers for intestinal inflammation in piglets based on their feeding behavior patterns during the peri-weaning period.** J. Suppi*[1], P. Salgado-López[1], E. Llauradó-Calero[1], C. Soldevila[2], A. Pelegrí-Pineda[3], Y. Saco[3], A. Bassols[3], and D. Solà-Oriol[1], [1]*Animal Nutrition and Welfare Service (SNIBA), Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain*, [2]*Vall Companys Group, 25191 Lleida, Spain*, [3]*Veterinary Clinical Biochemistry Service (SBCV), Department of Biochemistry and Molecular Biology, School of Veterinary, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain*. Weaning stress often leads to temporary feed deprivation, causing intestinal wall disruption and inflammation. Fecal biomarkers (fBM) such as calprotectin (fCal), lipocalin (LCN), myeloperoxidase (MPO), and adenosine deaminase (ADA) are used to detect inflammatory diseases in humans. The first two correlates with neutrophil counts, while MPO and ADA reflect immune cell activity. Little is known about how these BM behave in weaned piglets. We aimed to evaluate how these BM varies during the first days post-weaning (PW), in relation to piglets' feeding patterns. The trial involved 77 male piglets [(L×LW)×Pt], weaned at 21± 2.1 days (5.93 ± 0.05 kg). Dyed creep feed was provided peri-weaning to track intake via rectal swabs. Piglets were checked daily and fecal samples were collected during the first four days PW. Piglets were labelled as Creep-Feed Eaters (CFE; uninterrupted intake, n = 18), PW Eaters (PWE; started consuming PW, n d1 = 11, n d2 = 18, n d3 = 36), and Non-Eaters (NE; no feed intake, n = 23). FCal levels were analysed using an immunoturbidimetric method, LCN via ELISA, and ADA and MPO activities with colorimetric enzymatic assays. Data were analyzed using a linear mixed-effects model and ANOVA (R software). In the CFE group, both LCN and fCal remained stable over time ($P > 0.138$), while both MPO and ADA increased ($P < 0.003$) In the NE group, all fBM remained stable over days except MPO, which increased ($P = 0.006$). In the PWE group, LCN and fCal declined over time ($P < 0.007$), and MPO levels continued to rise ($P < 0.001$), while ADA remained constant ($P = 0.227$). Overall, the CFE group exhibited lower fCal and LCN levels but higher MPO and ADA levels than NE piglets ($P < 0.001$). LCN and fCal trends suggest that prolonged fasting translates into inflammation; while reintroducing the intake reduces it. ADA only varied in the CFE, suggesting a more mature immune response. MPO increased across all groups, indicating its release do not depend on feed intake. **Key Words:** feed intake, gut health, post-weaning diarrhea

## Theme V Posters: Mucosal Immunity and Pathogenesis and the Role of the Digestive Tract in the Maintenance of Health

**214 Gastrointestinal health and growth response to fiber supplementation and feeding regimens in grow-finish pigs.** N. A. Erker*, T. K. Everding, D. B. Paczosa, L. Meier, S. C. Fernando, P. S. Miller, and T. E. Burkey, *University of Nebraska - Lincoln, Lincoln, NE, USA*. The objective of the research was to assess the effects of fiber supplementation and time-restricted (TR) feeding on gastrointestinal (GI) health and performance of grow-finish pigs. Individually housed gilts (n=32, 94±1 d of age, 49±7kg BW) were used to test four treatment combinations with eight pigs per treatment. A 2x2 factorial design with two main factors was used: feeding regimen [ad libitum (ad lib) or TR] and fiber supplementation [no fiber supplement (NS) or fiber-supplemented with 0.5% Jerusalem Artichoke tubers and 25% soybean hulls (JA)]; resulting in four treatment groups: ad lib/NS (CTL), ad lib/JA (TRT1), TR/NS (TRT2), and TR/JA (TRT3). Diets consisted of a corn-soybean meal basal diet and the fiber supplemented diet replaced a combination of corn and soybean meal while maintaining consistent lysine levels. Pigs undergoing TR feeding had access to feed from 0615 – 1415 daily. Growth performance data was collected on d0, 14, 28, 42, 49, and 63. Blood and fecal samples were collected on d0, 28, 49, and 63. Microbial DNA from the fecal samples was prepped for 16S rRNA sequencing and data was processed through the DADA2 pipeline in R. PROC GLIMMIX in SAS was utilized to analyze the growth performance data and serum Intestinal Fatty Acid Binding Protein (I-FABP) levels. Overall, from d0 to 63 no effects were seen in growth performance. However, at d49 TR groups of pigs had reduced ADG compared to other treatments ($P = 0.052$) with improved G:F ($P < 0.0001$). These findings could have been influenced by a period of prolonged elevated environmental temperatures from d42-49. No significant differences were observed in microbial diversity between treatments; however, diversity was affected over time ($P < 0.001$). CTL had lower I-FABP than TRT1 ($P = 0.02$) on d49. In general, fiber supplementation and TR feeding neither negatively nor positively impacted growth performance or GI health. **Key Words:** fiber supplementation, time-restricted feeding, gastrointestinal health

**215 Oxidative stress and inflammation in pigs after challenge with *E. coli* lipopolysaccharide.** P. A. Madsen[1], D. Vodolazs'ka[1], M. S. Hedemann[1], A. R. Williams[2], and C. Lauridsen*[1], [1]*Aarhus University, AU Viborg, Denmark*, [2]*University of Copenhagen, Copenhagen, Denmark*. Infection with *E. coli* has been associated with increased inflammation and oxidative stress and is known to be the major cause of post-weaning diarrhea. This study validated biomarkers for systemic inflammation and oxidative stress providing insights into host immune responses. Ten healthy female pigs (3 months old, 25 ± 1 kg) were selected to ensure uniformity and avoid gender-related effects. A venous catheter enabled blood sampling at 0 (pre-LPS infusion), and 0.5, 1, 2, 3, 4, 6, 8, 12, 24, 36, 48 and 72 hours post-infusion. Two concentrations of LPS (low 0.75 and moderate 1.50 µg/kg BW) from *E. coli* O111:B4 were used. Changes in immunological and oxidative stress parameters were measured to assess temporal profiles and characterize immune and oxidative stress responses to the LPS challenge. Rectal temperature, plasma cytokines, acute-phase proteins, and oxidative stress markers were measured. Rectal temperature peaked three hours after LPS infusion (41.6 °C) and returned to pre-infusion levels within eight hours for both treatments (39.6 °C, $P \leq 0.03$). Plasma CRP levels increased post-LPS infusion, peaking at 12 hours (29.0 mg/L, $P = 0.001$), with no difference between LPS dosages ($P = 0.33$). Haptoglobin levels were generally higher in pigs receiving the moderate LPS dose than the low dose, particularly at 24 hours post-infusion (1.99 vs 1.28 mg/mL, $P = 0.01$). Pig-MAP levels peaked at 24 hours (0.72 g/L, $P < 0.001$) and were unaffected by LPS dose ($P = 0.62$). MDA levels peaked one hour after LPS infusion (2300 ng/mL, $P < 0.01$) and decreased within 12 hours to pre-infusion levels (950.5 ng/mL). The cytokines IL-6 and IFN-γ peaked 2 hours after LPS infusion, IL-10 after 1 hour, and IL-1β after 3 hours (1626, 10.1, 606 and 3.58 pg/mL, $P < 0.01$). In conclusion, MDA, acute phase proteins and inflammatory cytokines show distinct temporal patterns after LPS infusion and may function as biomarkers to evaluate interventions that modulate infection-induced oxidative stress. **Key Words:** LPS infusion, systemic

inflammation, biomarkers

**216 Evaluating Jerusalem Artichoke tubers as a prebiotic fiber source in weaned pigs: effects on growth performance and intestinal health.** N. A. Erker*, T. K. Everding, A. C. Neujahr, D. B. Paczosa, L. Meier, S. C. Fernando, P. S. Miller, and T. E. Burkey, *University of Nebraska - Lincoln, Lincoln, NE, USA*. The study aimed to assess the potential of Jerusalem Artichoke (JA) tubers as a source of prebiotic fiber in weaned pigs, focusing on their effects on growth performance and intestinal health. Nutritional research is an integral part of identifying alternatives to antimicrobials for maintaining herd health. Jerusalem Artichoke tubers have a relatively high level of inulin (~60%) as compared to other prebiotic fibers. Weaned pigs (n=64, 21d of age, 7.2±1.4kg BW) were used to test four dietary treatments with four pens per treatment. Treatments were randomly allocated to pens following a completely randomized design in a three-phase feeding trial. Diets consisted of a corn-soybean meal basal diet (control) and three treatment diets with ground JA tubers replacing corn at 0.05%, 0.5%, and 5.0% (JA-low, JA-mid, and JA-high, respectively) through all three phases. The trial ran from d0 to 35. Growth performance data was collected weekly. Blood samples were drawn weekly through d28 and fecal samples were collected on d0, 14, and 28. PROC GLIMMIX in SAS was utilized to analyze the growth performance and serum Intestinal Fatty Acid Binding Protein (I-FABP) data. Microbial DNA was extracted from the feces and prepped for 16S rRNA sequencing. The DADA2 pipeline in R was used to analyze the sequencing data. No significant difference was observed in growth performance or microbial diversity. On day 28, a significant difference ($P$ = 0.018) between I-FABP levels in the control and JA-high groups was noted and the relationship does not appear to be linear. Interpretation of the data was complicated by morbidity and mortality from *Trueperella pyogenes* that necessitated antibiotic treatment of the herd on d7. Despite these challenges, the results indicate that JA tubers can be safely included in weaned pig diets and may contribute to improved intestinal health when used at higher inclusion levels. Further research is needed to confirm these findings in larger, non-compromised herds. **Key Words:** prebiotic, intestinal health, weaned pigs

**217 Efficacy of 2, 4-dinitrobenzene sulfonic acid in the maintenance of a model of colitis in piglets.** Dominika Szkopek*[1], Jaroslaw Wolinski[1], Lukasz Kopiasz[2], Kamil Zaworski[1], Katarzyna Dziendzikowska[2], and Joanna Gromadzka-Ostrowska[2], [1]*Laboratory of Large Animal Models, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Instytucka 3, Jablonna, Poland*, [2]*Department of Dietetics, Institute of Human Nutrition Sciences, Warsaw University of Life Sciences, Nowoursynowska Str. 159C, 02-776 Warsaw, Poland*. The aim of the study was to evaluate the features of colitis in the domestic pig as an animal model of inflammatory bowel disease (IBD) in humans. The experiment was conducted on 16 male piglets, which were divided into 2 groups (n=8): control and colitis. After the adaptation period, colitis was induced by single rectal administration of DNBS solution (2, 4-dinitrobenzene sulfonic acid) in 50 % (v/v) ethanol at a dose of 80 mg/kg (1 ml) body weight. The concentration of DNBS was approximately 8.22% w/w. The final density of the ethanolic DNBS solution was approximately 0.977g/cm3. Animals in the control group were administered a 0.9% NaCl solution in the same volume in the same way. After 21 days, the animals were sacrificed, and a section of the colon was taken for histopathological examination (Geboes scale) and a section of small intestine for histomorphometry analysis, as well as blood for morphological analysis. Data were analyzed using ANOVA and Kruskal-Wallis tests, with normality checked via Shapiro-Wilk test. The blood morphological examination showed a significant increase in the relative neutrophil count in the control group ($P$ = 0.037), but these results were still within the hematological norms for the species. There were no significant differences in small intestine histomorphometry between groups ($P$ > 0.05). Histopathological examination of the colitis animals showed erosion and ulceration, which were absent in the control group, as well as increased chronic inflammatory infiltration and altered overall architecture compared to the control group. The results indicate that DNBS-induced colitis in the domestic pig may be a suitable research model for IBD. The absence of changes in blood

morphology and histomorphometry of the small intestine, with concomitant visible changes in histopathology of the colon indicates local inflammation, which was the aim of the present experiment. Study fund by National Science Centre (NCN), Poland. Grant number 2022/47/B/NZ9/03123. **Key Words:** colitis, pig model, Crohn's disease

**218 Time-course analysis of iron metabolism in the piglet model of iron deficiency and repletion after oral supplementation with Sucrosomial® Iron.** R.R. Starzynski[1], M. Lenartowicz[2], M. Ogluszka[1], G. Tarantino[3], E. Brilli[3], R. Mazgaj[1], Z. Kopec[1], X. Wang[1], B. Zelazowska[1], J. Wolinski*[4,5], D. Szkopek[4], and P. Lipinski[1], [1]*Department of Molecular Biology, Institute of Genetics and Animal Biotechnology, Polish Academy of Sciences, Jastrzebiec, Poland*, [2]*Department of Genetics and Evolutionism, Institute of Zoology and Biomedical Research, Jagiellonian University, Kraków, Poland*, [3]*Scientific Department, Pharmanutra S.p.A., Pisa, Italy*, [4]*Laboratory of Large Animal Models, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jablonna, Poland*, [5]*Department of Animal Physiology, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jablonna, Poland*. In pigs, iron deficiency (ID) is the most prevalent deficiency disorder occurring during the early postnatal period. Oral iron supplementation is the preferred treatment for ID. Sucrosomial® Iron (SI), a highly absorbable dietary iron supplement, a valid option for ID recovery. In this time-course study we examined iron fluctuations in the blood plasma, its tissue content/distribution, and the expression of iron metabolism genes following the administration of a single dose of SI to 14-day-old iron-deficient and iron-replete piglets (N=30). Data were analyzed using ANOVA followed by Duncan's multiple comparison tests for post hoc analysis. Results revealed essential differences in SI-derived iron pharmacokinetics between animals in the two groups. There was a more rapid increase in plasma iron levels and its accelerated clearance from the plasma of iron-starved piglets compared to that of the iron-sufficient animals, $P < 0.05$. The plasma hepcidin level, reached a maximum at 6 hours post-administration only in iron-replete piglets, 11,2 vs. 1,1 [ng/mL], $P < 0.01$. Accordingly, 24 hours after SI administration, levels of duodenal ferroportin, remained consistently low in these animals, whereas it displayed a significant increase in iron-deficient piglets, 1 vs. 1,68 [a.u.], $P < 0.05$. Consequently, staining for non-heme iron in the duodenum at this time point showed massive accumulation in the enterocytes of iron-replete piglets in the form of the iron-storage protein ferritin, 1 vs. 1,73 [a.u.], $P < 0.01$, suggesting increased iron retention. Results revealed fundamental differences in SI pharmacokinetics depending on the iron status and may be helpful in improving oral SI dosing schedules. Our findings emphasize the need to tailor the use of SI to the iron status of the individual animals and predict the involvement of hepcidin in the regulation of SI absorption. This study was supported by the National Science Centre, Poland, grant number 2020/39/B/NZ5/02469. **Key Words:** Sucrosomial® Iron, iron metabolism, iron deficiency

**219 Polyherbal mixture for sows associated or not to conventional anticoccidial treatment in the farrowing house: carry-over effects on the nursery piglets.** E. R. Oliveira*[1,6], A. P. P. Pavaneli[2,6], P. R. Gonçalves[3,6], F. Horta[2,4], C. Sol[4], and P. A. S. Rosa[5,6], [1]*Universidade Estadual de Londrina, Londrina, PR, Brazil*, [2]*Universidade de São Paulo, São Paulo, SP, Brazil*, [3]*Faculdade de Ciências Sociais e Agrárias de Itapeva, Itapeva, SP, Brazil*, [4]*Nuproxa, Ettoy, IA, Switzerland*, [5]*Centro Universitário do Cerrado Patrocínio, Patrocínio, MG, Brazil*, [6]*InsideSui, Patrocínio, MG, Brazil*. Anticoccidial treatment for piglets is a common practice for controlling coccidiosis during lactation, but pregnant sows are important agent carriers. This study aimed to evaluate the zootechnical performance of nursery piglets from sows supplemented with a polyherbal mixture (PHM, based on 15% saponins and 1.0-1.5% tannins) in combination with conventional anticoccidial treatment in the farrowing house. Forty-eight sows were divided into the treatments: Control (C1), no supplementation (n = 24); and Polyherbal (P), PHM from 90 days of gestation (500 g/ton) + lactation (300 g/ton; 23 days) (n = 24). Litters were distributed in the other two treatments (2×2 factorial): Control (C2), no anticoccidial treatment (n = 12); and Anticoccidial (A), toltrazuril 5% (1 ml/piglet) at 3 days old (n = 12). A total of 144 weaned piglets were followed up in the nursery phase according to the

previous experimental groups: C1+C2, P+C2, C1+A, and P+A. Feed intake was measured at 14, 28, and 42 days of nursery, and animals were individually weighed at 0, 14, 28, and 42 days of nursery. Parametric data were submitted to factorial analysis (DOE) while non-parametric data were evaluated by the Kruskal-Wallis test (Minitab 19). The PHM for sows in association with toltrazuril 5% for piglets resulted in the lowest feed conversion ratio (FCR) for the first 14 days (C1+C2 1.433$^{b}$, P+C2 1.611$^{a}$, C1+A 1.405$^{b}$, P+A 1.222$^{c}$, $P < 0.05$), and lower FCR values for the final phase (28-42 days of nursery) (C1+C2 1.453$^{ab}$, P+C2 1.489$^{a}$, C1+A 1.479$^{a}$, P+A 1.382$^{b}$, $P < 0.05$) and total period (C1+C2 1.403$^{b}$, P+C2 1.472$^{a}$, C1+A 1.412$^{b}$, P+A 1.372$^{b}$, $P < 0.05$). These results show a high synergy between the strategies, suggesting the polyherbal mixture for sows contributes to a better condition of the weaned piglet, improving its zootechnical performance in the nursery phase. **Key Words:** health natural diarrhea

**220 Feeding diets containing β-mannanase modulates immune response in growing-finishing pigs.** Y. H. de Paula*[1,2], G. M. Galli[3], C. J. Kippert[3], C. R. Oliveira[3], V. S. Cantarelli[1], L. Hauschild[4], M. Kipper[5], and I. Andretta[3], [1]*Federal University of Lavras, Lavras, Minas Gerais, Brazil*, [2]*University of Saskatchewan, Saskatoon, Saskatchewan, Canada*, [3]*Federal University of Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil*, [4]*São Paulo State University, Jaboticabal, São Paulo, Brazil*, [5]*Elanco Animal Health, São Paulo, São Paulo, Brazil*. Dietary β-mannanase (BM) can hydrolyze β-mannans, reducing the innate immune response induced by feeding glucomannanas. Thus, this study aimed to evaluate the effect of BM addition on the inflammatory response in grower-finisher pigs. Fifty-two non-castrated male pigs of a high-performance genotype (Landrace × Large White, 23.01 ± 0.415 kg) were assigned to two treatments: control diet (CON), a conventional phase-feeding program in which normal energy levels were used; and with BM, diets formulated with identical composition, except for a reduction of 35 kcal of net energy potentially compensated with BM addition (300 g/ton) during the whole 105 days of experiment. Blood samples were collected from seven pigs per treatment on days 21, 26, and 105 of the experiment, to analyze the cytokines IL-10 and TNF-α using an ELISA test kit. Fecal samples were collected directly from the rectal ampoule on the same days and in the same animals, to evaluate the biomarker calprotectin using an ELISA test kit. Statistical analyses were performed using the Tukey-Kramer Test in SAS Studio. Pigs fed with BM had expressively lower concentrations of IL-10 at 21 days (5.785 ± 4.765 pg/ml, $P = 0.005$), at 26 days (7.695 ± 7.695 pg/ml, $P = 0.011$), and at 105 days of experiment (17.75 ± 5.100 pg/ml, $P = 0.044$) when compared to CON diet (35.40 ± 4.765, 34.32 ± 7.695, and 38.05 ± 5.100 pg/ml, respectively). In addition, the BM also had lower concentration of TNF-α at 105 days (18.31 ± 10.16 pg/ml, $P = 0.003$) than the CON diet (86.24 ± 10.16 pg/ml), but there were no differences between the treatments at 21 and 26 days ($P > 0.05$). BM also decreased the calprotectin concentration at 105 days of experiment (3.475 ± 0.029 ng/ml, $P = 0.053$) when compared to CON diet (3.596 ± 0.029 ng/ml), but without differences at 21 and 26 days ($P > 0.05$). Therefore, the addition of β-mannanase reduces inflammatory responses associated with feeding in grower-finisher pigs. **Key Words:** Enzyme, Feed additive, Inflammation.

**221 Management of coccidial risk in sows: effects on the litters when natural polyherbal mixture is associated or not with conventional anticoccidial treatment in the farrowing house.** E. R. Oliveira*[1,4], A. P. P. Pavaneli[2,6], R. S. Oliveira[3,6], F. Horta[2,4], C. Sol[4], and F. G. D. Silva[5,6], [1]*Universidade Estadual de Londrina, Londrina, PR, Brazil*, [2]*Universidade de São Paulo, São Paulo, SP, Brazil*, [3]*Centro Universitário do Cerrado Patrocínio, Patrocínio, MG, Brazil*, [4]*Nuproxa, Ettoy, IA, Switzerland*, [5]*Universidade Federal de Viçosa, Viçosa, MG, Brazil*, [6]*InsideSui, Patrocínio, MG, Brazil*. Anticoccidial treatment for piglets is a common practice for controlling coccidiosis during lactation, but pregnant sows are important agent carriers. This study aimed to evaluate zootechnical performance and C*ystoisospora suis* oocyst count of litters from sows supplemented with a polyherbal mixture (PHM, based on 15% saponins and 1.0-1.5% tannins) in combination with conventional anticoccidial treatment for piglets in the farrowing house. Forty-eight sows were divided into the treatments:

Control (C1), no supplementation (n = 24); and Polyherbal (P), PHM from 90 days of gestation (500 g/ton) + lactation (300 g/ton; 23 days) (n = 24). Litters were distributed in the other two treatments (2×2 factorial): Control (C2), no anticoccidial treatment (n=12); and Anticoccidial (A), toltrazuril 5% (1 ml/piglet) at 3 days old (n = 12). *Cystoisospora suis* oocysts were quantified in piglet feces at 1, 5, 10, and 20 days old. Piglets were individually weighed at 2, 10, and 23 days old. Parametric data were submitted to factorial analysis (DOE) while non-parametric data were evaluated by the Kruskal-Wallis test (Minitab 19). The PHM alone or in association with toltrazuril 5% reduced the prevalence of animals positive for *Cystoisospora suis* (5 days: P 30% vs. C1 80%, $P < 0.05$; C1+C2 100%$^{a}$, P+C2 20%$^{b}$, C1+A 60%$^{ab}$, P+A 40%$^{ab}$, $P < 0.05$), (10 days: P 10% vs. C1 60%, $P < 0.05$; C1+C2 80%$^{a}$, P+C2 20%$^{b}$, C1+A 40%$^{ab}$, P+A 0%$^{b}$, $P < 0.05$), and (20 days: P 0% vs. C1 20%, $P < 0.10$). Both strategies also resulted in a lower coefficient of variation (piglet weight) at weaning (P 14.88% vs. C1 18.37%, $P < 0.05$; C1+C2 17.11$^{ab}$, P+C2 17.57$^{ab}$, C1+A 19.63$^{a}$, P+A 12.22$^{b}$, $P < 0.05$), while litters from polyherbal sows had less mortality before 10 days (P 1.238% vs. C1 3.290%, $P < 0.10$). These results suggest that alone or together with toltrazuril 5%, the PHM is an effective strategy to control coccidiosis in the farrowing house, reducing environmental contamination, piglet mortality, and litter heterogeneity at weaning. **Key Words:** health coccidiosis natural

**222 Influence of soybean-derived dietary trypsin inhibitor proteins on intestinal attachment of F18 enterotoxigenic *Escherichia coli* in weanling pigs subjected to experimental challenge.** M.J. Nisley*[1], E.R. Burrough[1], H.B. Krishnan[2], J.D. Spencer[3], O.F. Mendoza[4], and N.K. Gabler[1], [1]*Iowa State University, Ames, IA, USA*, [2]*University of Missouri, Columbia, MO, USA*, [3]*United Animal Health, Sheridan, IN, USA*, [4]*The Maschhoffs, Carlyle, IL, USA*. Our objective was to evaluate the extent to which dietary soybean-derived active TIU concentrations would exacerbate F18 enterotoxigenic *E. coli* (ETEC) disease-induced intestinal disruptions in weanling pigs. In a 3×2 factorial design, 288 weaned pigs (5.4 ± 0.92 kg body weight) were allocated by sex and FUT1 genotype across three dietary treatments (1.1, 2.4, or 4.2 TIU/mg complete feed) and two health conditions (control [CON]) or F18 ETEC (ETEC) challenged (8 pens/diet/challenge) over a 38-d period. After 10 d on diets, ETEC pigs were orally administered 5 ml of $3.8\times10^{9}$ cfu/mL F18 ETEC culture on day post-infection (dpi) 0. On dpi 5 and 12, one pig/pen was sacrificed to assess ileal mucosal attachment of F18 *E. coli* (*in situ* hybridization). Pooled pen feces were collected on dpi 0, 3, 5, 7, 9, 11, 14, and 21 to assess dry matter (DM) and nitrogen digestibility (dpi 14 to 21). Pen was the experimental unit and data were analyzed for main effects and interaction between diet and ETEC challenge. Increasing dietary TIU reduced pig weight gain and feed intake by up to 15% ($P < 0.05$), while ETEC2.4 had higher mortality incidence than CON2.4 (20.8 vs 0%, $P < 0.01$). No interaction was observed for ileal F18 attachment on dpi 5 or 12 ($P > 0.05$). On dpi 5, ileal F18 attachment was increased in ETEC versus CON pigs ($3.7\times10^{-3}$ vs $0.1\times10^{-3}$ F18 copies/μm$^2$, $P < 0.0001$). By dpi 12, ileal F18 attachment did not differ between challenge groups ($P > 0.05$), suggesting disease resolution. Fecal DM was reduced in ETEC pigs ($P < 0.05$), but no interaction between diet, ETEC, and day were observed ($P > 0.05$). The apparent total tract digestibility of nitrogen from dpi 14 to 21 was indifferent ($P > 0.05$). On dpi 5, ETEC pigs had reduced luminal butyric acid, propionic acid, valeric acid, aspartic acid, and glutamic acid concentrations ($P < 0.01$). In conclusion, increasing TIU concentration did not affect F18 ETEC attachment but reduced pig growth and increased mortality incidence in challenged pigs. **Key Words:** ETEC, trypsin, weanling

**223 Supporting health of newly weaned pigs: the role of a microencapsulated blend of botanicals during an F18$^{+}$ *Escherichia coli* challenge.** Andrea Bonetti*[1], Yesid Garavito-Duarte[2], Benedetta Tugnoli[1], Hyunjun Choi[2], Andrea Piva[1,3], Ester Grilli[3,4], and Sung Woo Kim[2], [1]*Vetagro S.p.A., Reggio Emilia, 42124, Italy*, [2]*Department of Animal Science, North Carolina State University, Raleigh, NC 27695, USA*, [3]*Department of Veterinary Medical Sciences, University of Bologna, Ozzano dell'Emilia, 40064, Italy*, [4]*Vetagro Inc., Chicago, IL 60603, USA*. The aim of this study was to evaluate the efficacy of a microencapsulated blend of botanicals (BOT) in diets for newly weaned pigs orally challenged with F18$^{+}$ enterotoxigenic *E.*

*coli* (ETEC). Sixty-four 21-day-old weaning pigs were divided in 4 groups (n = 16): negative control (NC, basal diet); positive control (PC, basal diet); treated groups with BOT added in basal diet (BOT1: 1 kg/MT; BOT2: 2 kg/MT). On d7, PC, BOT1, and BOT2 were challenged with ETEC ($1.5\times10^{10}$ CFU). Growth performance and fecal score were recorded until d28. On d14, half of pigs were euthanized to collect jejunal samples for mucosa-associated microbiota and intestinal health analysis. Data were analyzed with SAS MIXED procedure. During d7 to 14, PC had lower ($P < 0.05$) growth performance compared to NC. During d7 to 28, G:F was improved ($P < 0.05$) in BOT1 and BOT2 compared to NC and PC. From d7 to d11, fecal score was worsened ($P < 0.05$) in PC compared to NC but, on d10, BOT improved ($P < 0.05$) it compared to PC. Despite the challenge, the mucosa-associated microbiota of PC was not different from NC, but BOT1 and BOT2 showed lower ($P < 0.05$) *Staphylococcus* compared to NC and PC. However, PC had reduced ($P < 0.05$) ZO-1 and occludin in the jejunum compared to NC, whereas BOT1 and BOT2 increased ($P < 0.05$) their expression and tended to reduce TLR4 levels compared to NC and PC. The jejunal mucosa of PC also had higher ($P < 0.05$) IL-6, but BOT1 and BOT2 reduced its concentration to the same of NC. Moreover, intestinal Ki-$67^{+}$ was reduced ($P < 0.05$) in PC, but it was increased ($P < 0.05$) in BOT1 and BOT2 compared to PC. In conclusion, BOT could improve growth performance and fecal scores of newly weaned pigs by supporting their health during ETEC challenge through the modulation of mucosa-associated microbiota, reduction of inflammatory response, and enhancement of gut integrity in the jejunum. **Key Words:** Pigs, Enterotoxigenic Escherichia coli, Botanicals

**224 Galacto-oligosaccharides can prevent enterotoxigenic *Escherichia coli* adhesion and intestinal injury *in vitro*.** S. Tanghe*[1], B. Guantario[2], A. Finamore[2], C. Devirgiliis[2], S. Verstringe[1], M. De Vos[1], J. Vande Ginste[1], and M. Roselli[2], [1]*Agrifirm, Drongen, Belgium*, [2]*CREA-Research Centre for Food and Nutrition, Rome, Italy*. Post-weaning diarrhea in piglets is frequently caused by enterotoxigenic *Escherichia coli* (ETEC). The objective of this *in vitro* study was to examine the possible protective effect of galacto-oligosaccharides (GOS) on ETEC-induced intestinal injury. Growth inhibition of ETEC F4 in the presence of GOS was assessed through OD measurement of ETEC cultures. The ability of GOS to reduce ETEC F4 adhesion was tested in the intestinal Caco-2 cell line. Cell permeability was assayed by measuring the transepithelial electrical resistance (TEER) and phenol red apparent permeability (Papp) in Caco-2 cells treated with GOS in the presence or absence of ETEC F4, to evaluate if GOS was able to protect intestinal cells against ETEC-induced membrane damage. Phenol red Papp values below $1 \times 10^{-6}$ cm $s^{-1}$ were considered indicative of intact monolayers and set as threshold value, irrespectively of statistical significance among samples. Phosphorylated-p65/p65 protein expression ratio was analyzed by Western Blot to measure if GOS could counteract ETEC-induced NF-κB activation, leading to inflammation. Tests were performed with 2% GOS. No direct antibacterial effect of GOS against ETEC F4 was observed in the growth inhibition assay. Adhesion of ETEC F4 to Caco-2 intestinal cells was reduced by GOS (6.5 vs 5.9 log CFU/mL, for ETEC vs ETEC+GOS; $P < 0.001$). No significant effect of GOS on TEER in the presence of ETEC F4 was observed, but Papp restored below the threshold value in the presence of GOS ($2.2\times10^{-6}$ vs $8.7\times10^{-7}$ cm $s^{-1}$, for ETEC vs ETEC+GOS). Protein expression of phosphorylated-p65/p65 was reduced by GOS (2.76 vs 0.81, ETEC vs ETEC+GOS; $P = 0.001$), indicating lower ETEC-induced NF-κB activation. These *in vitro* results demonstrated that GOS can prevent ETEC adhesion, protect intestinal cells from ETEC-induced membrane damage, and counteract ETEC-induced NF-κB activation, which may indicate the potential of including GOS in piglet weaner diets to prevent ETEC-induced intestinal injury. **Key Words:** galacto-oligosaccharides, enterotoxigenic *Escherichia coli*, post-weaning diarrhea

**225 Effects of fecal microbiota transplantation from domestic pigs on oxidative stress and immunity in weaned piglets challenged with lipopolysaccharides.** M. A. K. Azad*[1,2], G. Gao[1], Q. Zhu[1,2], B. Qin[1,2], and X. Kong[1,2], [1]*Institute of Subtropical Agriculture, Chinese Academy of Sciences, Changsha, Hunan, China*, [2]*College of Advanced Agricultural Sciences, University of Chinese Academy of Sciences, Beijing, China*. Domestic pigs have higher resistance to weaning stress

due to their adaptability to the local environment and distinct microbiome/mycobiome characteristics. Thus, this study explored the impacts of FMT from domestic pigs (Taoyuan Black) on oxidative stress and immunity in commercial (DLY; Duroc × Landrace × Yorkshire) piglets using a LPS-challenge model. Twenty-eight piglets (DLY; half-male and half-female) were allocated into four experimental groups using a 2×2 factorial arrangement in a completely randomized design with the effects of FMT and LPS challenge. The experimental groups consisted of a basal diet with intramuscular injection of saline, a basal diet with oral gavage FMT (1.50 mL/d), a basal diet with LPS challenge, and a basal diet with oral gavage FMT (1.50 mL/d) with LPS challenge. The feeding experiment lasted 28 d. Intraperitoneal injections of LPS 100 μg/kg BW or equal amounts of sterile saline were given to experimental piglets on d 28. After 4 h of the LPS challenge, all piglets were selected and euthanized for sample collection and analyses. Blood samples were drawn from the anterior vena cava into heparin sodium-containing tubes, centrifuged at 3000×*g* for 10 min to obtain plasma for further analyses. The jejunal and ileal mucosa samples were collected and immediately stored at -80°C for further analyses. The results showed that FMT inhibited the increase of plasma diamine oxidase ($P < 0.05$) induced by LPS. Moreover, FMT treatment inhibited ($P < 0.05$) the decrease of GSH-Px and T-OAC and the increase of MDA levels in plasma induced by LPS. The concentrations of intestinal immuno-cytokines, including IL-1β, IL-6, IL-10, TNF-α, and IFN-γ, were increased ($P < 0.05$) by FMT treatment in the jejunum of piglets. Furthermore, FMT treatment inhibited the increase of TNF-α and IFN-γ concentrations in the jejunum of piglets induced by LPS ($P < 0.05$). In conclusion, FMT from domestic pigs is effective to improve the host immunity in LPS-challenged piglets. **Key Words:** FMT, piglets, immunity

**226 *Lactobacillus* postbiotics improved postweaning piglets' growth performance and diarrhea situation under E.coli challenge conditions.** Yanhong Luo*[1,2], Stephane Duval[2], Maria Walsh[2], and Philippe Tacon[3], [1]*dsm-firmenich Nutritional Products, Animal Nutrition Research Center, Bazhou, Hebei, China*, [2]*dsm-firmenich Nutritional Products, Kaiseraugst, Aargau, Switzerland*, [3]*dsm-firmenich Houdan, Route de Bû, Houdan, France*. Weaning is one of the most stressful events in pig's whole life and the swine industry continually seeks effective solutions to support piglets during this period. Postbiotic have recently attracted attention from researchers due to their ability to improve host gut health, along to their safety, stability and easy to use. Therefore, this study was designed to explore the effect of *Lactobacillus* postbiotics on postweaning piglets growth performance and health under *E.coli* challenge condition. A total of 336 piglets (21d old) were used and distributed into 6 treatments with 14 replicates per treatment and 4 piglets per pen. The trial used a 3X2 factor design: 3 control diets and 2 postbiotics doses (0, 20ppm). The 3 controls included a negative control group (NC), and the NC added with 2000ppm ZnO (PC1) or ZnO and 440 ppm CTC (PC2) during d0-22. All the pigs were orally provided with 5mL *E. coli* K88 ($10^{\wedge 9}$ CFU/mL) at d10. The trials lasted for 42 days, during which growth items was recorded. Clinical and fecal scores were measured at d10-28. The analysis on the main effect of control and postbiotics diets showed no correlation between them, and the postbiotics significantly improved BW, ADG and FCR after challenge (d10-22) and reduced the clinical and diarrhea scores ($P < 0.01$), and showed a trend to improve BW, ADG and FCR at d0-10 ($P < 0.05$). Multiple comparison revealed that postbiotics added in the NC diet improved ADG and FCR at d0-22 and decreased the clinical and diarrhea scores ($P < 0.05$), achieving similar efficacy with PC1&PC2 groups. In addition, the postbiotics added in the PC1 diet further increased the BW and ADG at d0-22 and reduced the clinical and diarrhea score ($P < 0.05$). In conclusion, the postbiotics added in the diet improved growth performance and reduced diarrhea and abnormal clinical issues in piglets under *E.coli* challenge condition, and achieved similar efficacy with PC groups, making it a high potential solution for supporting postweaning piglets. **Key Words:** Postbiotics, E.coli challenge, weaning piglets

**227 Maternal Hydroxy-Selenomethionine Supplementation During Pregnancy and Lactation Enhances Offspring Performance by Improving Intestinal Morphology and Redox Status.** J. Wang[1,2], H. Hua[1,2], Z. Peng[1,2], S.Q. Wang[1,2], M.A. Hachemi[3], D. Bloxham[3], D. Cardoso*[3], B. Mallmann[3], and L.H. Sun[1,2], [1]*Department of Animal Nutrition and Feed Science, HZAU, Wuhan, Hubei, China*, [2]*Hubei Hongshan Laboratory, Wuhan, Hubei, China*, [3]*Adisseo France S.A.S., Antony, France*. This study evaluated the benefits of hydroxy-selenomethionine (OH-SeMet) supplementation in sows during gestation and lactation in summer, compared to seleno-yeast (SY) and sodium selenite (SS), on the performance, redox status, and gut morphology of their offspring. A total of 60 sows of the same parity (Landrace × Yorkshire) with similar body weights were randomly assigned to three groups (n = 20/group): basal diet (BD; with Se ≤ 0.1 mg/kg) + 0.3 mg Se/kg as SS, BD + 0.3 mg Se/kg as SY, or BD + 0.3 mg Se/kg as OH-SeMet. The trial was conducted from gestation to weaning at day 21 at a commercial experimental farm during summer (THI > 75). Ten piglets per treatment were euthanized at weaning to collect small intestinal samples for gut morphology, selenoprotein expression, and redox status analyses. Data were analyzed using one-way ANOVA with significance set at $P < 0.05$. Compared to SS and/or SY, maternal OH-SeMet improved piglets growth performance, as indicated by a higher number of piglets weaned at day 21 (11.5 for OH-SeMet vs 10.2 and 10.5 respectively for SY and SS; $P \leq 0.1$) and greater litter body weight at day 21 (77.6 kg for OH-SeMet vs 62.2 and 64.1 kg for SY and SS respectively; $P \leq 0.05$). Morphological results showed that OH-SeMet reduced crypt depth and increased villus-to-crypt ratios in the ileum ($P \leq 0.05$) and exhibited a trend toward reduced crypt depth and increased villus-to-crypt ratios in the duodenum and jejunum ($P \leq 0.10$), with SY showing intermediate values. OH-SeMet enhanced selenoprotein N and glutathione peroxidase 4 protein gene expression in the jejunum compared to SS and SY ($P \leq 0.05$). OH-SeMet increased total antioxidant capacity compared to SS, with SY showing intermediate values ($P \leq 0.05$). Supplementing sows with OH-SeMet during gestation and lactation specially under heat stress conditions could be an effective strategy to enhance redox status, prevent gut health issues in neonatal piglets, and ensure optimized growth performance. **Key Words:** selenium hydroxy-selenomethionine, antioxidant

**228 Impact of a Phytogenic Feed Additive on Diarrhea Incidence, Intestinal Histomorphology and Fecal Excretion of F4-Fimbriated Enterotoxigenic *Escherichia coli* in Postweaning Piglets.** Alberto Torres-Pitarch*[1], Anja Keiner[1], Maud Le Gall[1], Francesc Molist[2], Guan Xiaonan[2], Anouschka Middelkoop[2], Encarnacion Jimenez-Moreno[1], Aitor Balfagon[1], Graziano Mantovani[1], Miquel Nofrarias[3], and Tobias Aumiller[1], [1]*Cargill Animal Nutrition and Health, Schiphol, Netherlands*, [2]*Schothorst Feed Research, Lelystad, Netherlands*, [3]*IRTA. Animal Health Program. Centre de Recerca en Sanitat Animal (CReSA), Bellaterra, Spain*. The study investigated the effects of a phytogenic feed additive (PFA) containing herbs, plant extracts and essential oils from *Lamiaceae, Schisandraceae, Zingiberaceae* and *Fabaceae* families on fecal score, intestinal histomorphology and fecal excretion of F4-fimbriated enterotoxigenic *Escherichia coli* (F4-ETEC) in post-weaning piglets. Thirty weaned piglets [Tempo x TN70 (Large White x Norsvin Landrace), 30.7 ± 0.70 days old, 9.4 ± 1.28 kg (mean ± SEM)] were randomly allocated to three treatment groups. The positive control (PC) group received colistin via drinking water from days 8-14 post-weaning and the same basal diet as the negative control group (NC); the treatment group (PFA) received the basal diet with PFA supplementation. The experiment lasted 21 days. At day 9 post-weaning, all piglets were orally administered 3.0x10^10 CFU/piglet of the F4-ETEC strain. The experimental data was analyzed by ANOVA using GenStat® version 23. Multiple comparisons between treatments groups were tested with Tukey adjustment. Positive Control piglets had higher fecal consistency than NC and PFA piglets (Respectively, 6.4 *vs* 5.7 *vs* 5.9 ± 0.13, $P < 0.05$) during the entire study. PFA supplementation resulted in a lower percentage of piglets excreting F4-ETEC in the feces on d4-7 post-challenge than in the NC group ($P < 0.05$), but a higher percentage versus the PC group on d3-7 post-challenge ($P < 0.05$). The number of goblet cells (GC) in the jejunum of PFA piglets was higher than the NC and PC piglets (Respectively, 7.4 *vs* 5.0 *vs* 3.7 ± 0.68 GC/villi, $P < 0.01$). Globet cells density in the jejunum of PFA piglets was larger than in PC piglets (1.37 *vs* 0.69 ± 0.186

GC/µm, $P < 0.05$) and similar to NC piglets ($P > 0.10$). Mucus thickness in the jejunum and colon of PFA piglets was similar to NC piglets and PC piglets ($P > 0.10$). In conclusion, PFA supplementation to F4-ETEC challenged piglets reduced *E. coli* excretion in feces and improved jejunal histomorphology. **Key Words:** piglet, *E.coli*, phytogenics

## Theme IV: Development of Digestive and Absorptive Capacity in the Neonate and Impact of Weaning on Intestinal Function

**229 Use of organoids to study the role of the microbiota in the early life development of the pig intestine.** M. Beaumont*, *GenPhySE, Université de Toulouse, INRAE, ENVT, Castanet-Tolosan, France*. The gut microbiota plays a key role in the postnatal development of the pig intestine. To understand how gut microbes contribute to the maturation of the epithelial barrier, we use pig intestinal organoids. Porcine intestinal organoids consist of a single layer of epithelial cells that self-organize in 3D and contain both absorptive and secretory cells, all derived from stem cells. We performed organoid experiments to investigate whether the early life microbiota imprints intestinal stem cells, which could influence long-term gut health. Our results show that organoids derived from piglets depleted of dominant Gram-negative bacteria (e.g. Enterobacteriaceae) during the first week of life expressed lower levels of innate immunity genes, indicating intestinal stem cell imprinting. In contrast, intestinal organoids derived from suckling or weaned piglets had a similar phenotype, despite major differences in the *in vivo* microbiota. These results suggest that the imprinting of intestinal stem cells by the gut microbiota may be restricted to the period immediately after birth. We also developed the culture of cell monolayers derived from porcine intestinal organoids in order to facilitate the access to the apical side of epithelial cells where interactions with the gut microbiota occur. Experiments with this 2D model showed that the gut microbiota-derived metabolites butyrate and isovalerate improved epithelial barrier function. We also demonstrated in this model that butyrate was able to reduce epithelial barrier dysfunction induced by the foodborne mycotoxin deoxynivalenol. Taken together, our results highlight the interest of using porcine intestinal organoids to elucidate how the gut microbiota influences the epithelial barrier function. Ongoing complexification of organoid cell culture by using gut-on-chip systems will allow us to more closely model the pig intestine *in vitro*. **Key Words:** enteroids, epithelial barrier, bacterial metabolites

**230 Maternal dietary live yeast supplementation alters jejunal mucosal proteomes of piglets during suckling and postweaning phases.** Yuechi Fu*[1], Theresa Casey[1], Timothy Johnson[1], Jun Xie[2], Olayiwola Adeola[1], and Kolapo Ajuwon[1], [1]*Department of Animal Sciences, Purdue University, West Lafayette, IN 47907, United States*, [2]*Department of Statistics, Purdue University, West Lafayette, IN 47907, United States*. Maternal dietary live yeast (LY) supplementation has been shown to enhance intestinal development of offspring, however, the mechanisms underlying maternal LY-induced programming of piglet intestinal development remain unclear. This study investigated changes in jejunal mucosal proteomes of piglets in response to maternal dietary LY supplementation during the suckling and early postweaning periods. On d 77 of gestation, 20 sows were assigned to each of 2 dietary treatments: without (CON) or with LY (*Saccharomyces cerevisiae*) supplementation at 0.05% of diet during gestation and 0.1% during lactation. Jejunal mucosa was collected from 6 pigs in each treatment during suckling (postnatal day (PND) 0, 10, 18) and the early postweaning stages (postweaning day (PWD) 7 and 14). Total proteins were extracted from mucosal samples and subjected to liquid chromatography-tandem mass spectrometry for shotgun proteomic analysis. Data were analyzed using Welch's t-test to compare differences between piglets from CON and LY sows in RStudio (version 4.3.1). Significance was set at raw $P < 0.05$. A total of 23, 50, 75, 66, and 54 differentially abundant proteins (DAPs) were identified in piglets from CON and LY sows on PND 0, PND 10, PND 18, PWD 7, and PWD 14, respectively ($P < 0.05$). Gene ontology enrichment analysis during the suckling period showed that DAPs were enriched in pathways associated with chemical synaptic transmission, regulation of mitochondrial membrane potential, glycogen catabolic, and biosynthetic processes. The DAPs were primarily involved in carbohydrate phosphorylation, muscle contraction, positive regulation of immunoglobulin production, and non-canonical nuclear factor kappa B signaling transduction ($P < 0.05$) after weaning. Taken together, the findings suggest potential benefits of maternal dietary LY supplementation in modulating mucosal energy metabolism, gut barrier function, and immune development during early piglet

growth. **Key Words:** early development, jejunal mucosal proteome, maternal transfer

**231 Hypothalamic and ileal transcriptomic insights of poorly adapted freshly weaned pigs.** L. Fabà*, T. G. Hulshof, M. O Wellington, and H. M. J. Van Hees, *Trouw Nutrition R&D, Swine Research Centre, Boxmeer, The 8 Netherlands*. The objective of this study was to describe the hypothalamic and ileal transcriptomic profile in poor performing pigs at 1-week postweaning. Four pens (3 pigs each) were chosen from 40 based on high or low feed intake from day 0 to day 3 (weaning weight ~5.1 kg; N = 12 pigs). Pigs were dissected on day 7 and reclassified by average daily gain (ADG) as high (242 g/d) or low (98 g/d) to refine grouping (one swap) with individual performance (n = 6 pigs each). Ileal tissue, collected in RNAlater, and hypothalamus, snap-frozen at -80°C, were sent for RNA extraction and deep mRNA sequencing (Novogene, UK). Quantitative pathway activation was analyzed using SAVVY™ pathway catalogues (BIOFRACTAL, Portugal). Differential gene expression was analyzed by ranking DeSeq2 normalized genes. All results reported here are logarithmic fold changes with $P < 0.05$. In the ileum, low-ADG pigs showed inhibited barrier function, absorption of lipids, carbohydrates, and osmolytes. The metabolism of lipids, vitamins, and some amino acids was downregulated in low ADG pigs. Similarly, pro-inflammatory response, leukocyte migration, neutrophil degranulation, and antimicrobial peptides were inhibited, while the complement cascade was upregulated. In the hypothalamus, low ADG pigs showed inhibited MHCII and T-cell signaling but had upregulated complement cascade, several pro-inflammatory cytokines, and toll-like receptors. Low ADG pigs' hypothalamus showed inhibited antioxidant capacity and cellular stress (i.e., HIF1A gene associated with hypoxia stress). They had upregulated mTOR1 and GLP1 pathways which suppress appetite, and upregulated thermogenesis and mitochondrial biogenesis. In conclusion, low ADG pigs had intestinal disruption, a weakened ileal immune response and elevated complement cascade, plus hypothalamic inflammation, metabolic stress, and suppressed appetite at 1-week postweaning. Hence, this study offers new insights into the gut-brain adaptation in postweaning pigs. **Key Words:** Gut-brain axis, Feeding, Weaning

**232 Comparation of intestinal development of different pigs reveals PPARα is involve in regulating intestinal villus size and nutrients digestibility.** Q. Wang*[1], L. Yin[1], Z. Wang[1], J. Li[1], Q. Wang[1], J. Li[1], Y. Yin[2], and H. Yang[1,2], [1]*College of Life Sciences, Hunan Normal University, Changsha, Hunan 410081, China*, [2]*Institute of Subtropical Agriculture, Chinese Academy of Sciences, Changsha, Hunan, 410125, China*. Intestinal morphology and epithelial surface change in response to the degree of breed inheritance and environmental conditions, but more work is needed to understand the regulatory mechanisms by which this occurs. Here, we selected six each of Shaziling and Yorkshire pigs at 30, 60, 90, 150, 210 and 300 days of age to compare their intestinal development. The results showed that the length and weight of the small intestine of Yorkshire pigs were significantly greater than those of Shaziling pigs ($P < 0.05$), villus height in different intestinal segments of Shaziling was greater than that of Yorkshire pigs ($P < 0.05$). The number of Ki-67 cells in the jejunal crypts and the number of goblet cells were also significantly greater in Shaziling pigs than in Yorkshire pigs ($P < 0.05$). At 90 days of age, the activities of alkaline phosphatase and disaccharidase in Shaziling pigs and were significantly greater than those in Yorkshire pigs ($P < 0.05$). Jejunal mRNA abundance of nutrient transporters in Shaziling pigs was significantly higher than those in Yorkshire pigs ($P < 0.05$). Transcriptomic sequencing showed that the main differences between the two were focused on lipid metabolism, including the PPARα signaling pathway. The addition of GW6471, an inhibitor of PPARα, inhibited the budding and growth of intestinal organoid ($P < 0.05$), whereas the agonists WY14643 and N-oleoyl ethanolamine (OEA) both promoted the growth of organoid ($P < 0.05$). Moreover, OEA promotes cell proliferation and budding of organoids by enhancing oxidative phosphorylation through PPARα. Dietary OEA improves intestinal morphology and nutrient digestion and absorption by promoting the proliferation and differentiation of intestinal stem cells in piglets. Taken together, this study demonstrates the different intestinal developmental patterns of Shaziling and Yorkshire pigs and provides a potential target for

improving digestive and absorptive efficiency by increasing villus size. **Key Words:** Villus height, intestinal functions, PPARα

**233 Multi-omics characterization of swine colostrum and association of bioactive compounds with piglet survival and growth.** F. Correa[1], G. Rocchetti[2], P. Trevisi[1], M. Errico[2], B. Polimeni[1], A. Serra[3], M. Mele[3], L. Lucini[2], A. Gallo[2], and D. Luise*[1], [1]*Department of Agricultural and Food Sciences (DISTAL), University of Bologna, Bologna, Italy*, [2]*Department for Sustainable Food Process, Universita ` Cattolica del Sacro Cuore, Piacenza, Italy*, [3]*Department of Agricultural, Food and Agro-Environmental Sciences, University of Pisa, Pisa, Italy*. Colostrum is recognized as the "golden elixir of health" due to its optimal chemical, immunological and nutraceutical properties for newborns, but little is known on swine colostrum. This study aims to provide a multi-omics characterization of sow colostrum to identify the most relevant bioactive compounds associated with piglet survival and growth, with the ultimate goal of improving artificial colostrum formula. 20 samples of colostrum and individual piglet performance data were collected at farrowing (d0), day 7 (d7) and 24 (d24). Colostrum was analyzed for proximal composition, metabolomic and lipidomic profile. Litters were classified into survival (d0-d7:<12% or >12%; d0-d24:>18 or >18%) and average daily gain (ADG, g/d) classes (d0-d7: <140; 140-200;>200; d0-d24:<200;200-240;>240). Data were analyzed using a multivariate OPLS-DA and sPLS-DA approach. 950 metabolites (108 confirmed) and 72 fatty acids were characterized in colostrum. Colostrum composition allowed to correct clustering for piglet mortality from d0-d7 ($Q^2$>0.50). A total of 177 metabolites and 5 fatty acids discriminated for high survivability (<12% mortality); alpha-TOCHOPHERYL ACETATE, monolinolein, Cholest-4,6-Dien-3-One (3-hydroxyhexadecanoyl)glycine (Vip scores: 3.50, 3.24, 3.39, 2.10) and CLA 8t10c (sPLA-DA score 1) discriminated for the higher survivability. A total of 78 metabolites and 60 fatty acids discriminated for ADG d0-d24; alpha-TOCHOPHERYL ACETATE, Docosapentaenoic acid and Surugamide A (VIP score 3.4, 2.23,2.38) and C23:1c14, C16:1t9, C15 ante fatty acids (sPLA-DA score 0.32, 0.16, 0.15) discriminated for the higher ADG (ADG > 240 g/d). In conclusion, this study suggests the presence of several bioactive compounds in colostrum that may be associated with improved survival and growth of piglets and with the maturation of the gut microbiota. The results of the present study may also be of interest for the development of future dietary supplements of artificial colostrum. **Key Words:** newborns, vitamin E, conjugated linoleic acid

**234 Investigating the Impact of Early-Life Gut Microbiota Perturbation on Porcine Physiology and Immune Function.** L. Comer, M. Z. Akram, H. Zhao, and N. Everaert*, *Nutrition and Animal Microbiota Ecosystems Lab, Department of Biosystems, KU Leuven, Heverlee, Belgium*. Early life represents a detrimental window in the shaping of the immune system by the gut microbiota, yet this delicate interplay can be perturbed by factors such as weaning and antibiotic use. We sought to simulate two contrasting rearing conditions with varying levels of microbiota disruption, and as such, aimed to elucidate their longer-term microbiota and immunological impacts. A total of 32 piglets (Piétrain × Belgian Landrace) were divided equally into two groups from farrowing: compromised piglets received antibiotics (intramuscular trimethoprim-sulfamethoxazole and oral amoxicillin) in the first week of life, were weaned at 3 weeks and had no bedding material; optimal piglets received no antibiotics, were weaned at 6 weeks and had bedding material (straw and wood shavings) throughout. Diet remained identical for both groups. The spleen, ileum, faeces and blood were taken at day 90. Bodyweight was measured throughout. The microbiota was sequenced via PacBio V1-V9 16S sequencing, while gene expression was analysed via high-throughput qPCR. The optimal group showed significantly increased bodyweights on days 27 and 90 (Wilcoxon $P < 0.05$). The faecal microbiota β-diversity was significantly different (PERMANOVA $P < 0.05$) at day 90, with optimal pigs exhibiting increased *Butyricoccaceae UCG-008* while the compromised group showed increased lactic-acid-producing bacteria such as *Lactobacillus* (LEfSe LDA>3). Gene expression of the pattern recognition receptor genes *NOD2*, *CLEC7A* (spleen), and *TLR4* (ileum) was significantly increased among the optimal pigs (Wilcoxon $P > 0.05$). On the contrary, compromised pigs showed

increased expression of the inflammasome gene *NLRP3* (ileum). Increased neutrophil reactivity and granularity intensity was also observed in compromised pigs in the blood ($P < 0.05$). These data demonstrate that early-life perturbations in the microbiota can exert lasting physiological and immunological impacts on the host. **Key Words:** Microbiota, weaning, immunology

**235 Sensory additive in creep feed modulates post-weaning immune development and metabolism in piglets.** Z.W. Ng'ang'a[1,2], N. Tous[1], J. Tarradas[1], R. Beltrán-Debón[2], J.J. Pastor[3], S. López-Vergé[3], G. Tedo[3], and D. Torrallardona*[1], [1]*IRTA, Animal Nutrition, Constantí, Catalonia, Spain*, [2]*Universitat Rovira i Virgili, Tarragona, Catalonia, Spain*, [3]*Lucta S.A., Cerdanyola del Vallès, Barcelona, Spain*. Early nutritional interventions play a crucial role in shaping gut immune development and overall health in weaned piglets. In this study, we examined the impact of sensory additives in creep feed on post-weaning gene expression associated with immune response and digestive function in piglets. Twelve sows were split into 2 groups and their litters were offered either a control (T1) creep feed or a creep feed with sensory additive (T2) from 7d of age. At weaning, 6 eaters and 6 non-eaters per group (total 24) were selected based on the presence of indigo-carmine feed marker in rectal swabs for 3 days prior to weaning. At d5 PW, they were sampled to test intestinal immune development. The expression of a panel of genes involved in immune response, oxidative stress and metabolism was evaluated by high-throughput microfluidic qPCR in jejunal and ileal mucosa. Eater category significantly upregulated the expression of *IL10* ($P = 0.04$) and *DEFB1* ($P = 0.02$) in jejunum, suggesting enhanced anti-inflammatory and antimicrobial responses, respectively. T2 upregulated the expression of *DAO* ($P = 0.03$), *PPARGC1A* ($P = 0.02$), and *ALPI* ($P = 0.03$) in ileum, enhancing digestive enzyme activity, metabolic regulation and mucosal defense. Interactions between treatment and eating category were observed for *PPARGC1A* ($P = 0.04$), *GPX2* ($P = 0.02$) and *NFKBIA* ($P = 0.05$) in jejunum, and *TLR2* ($P = 0.03$) and *FAXDC2* ($P = 0.04$) in ileum, implying the T2 modulation of inflammatory, metabolic and antioxidant pathways in creep feed eaters. Interaction tendencies were also observed for *IL8*, *IL10* and *CCK* in jejunum and *IL8*, *IFNGR1*, *HNMT* and *NFKB1* ($P < 0.1$) in ileum. Overall, these results indicate that pre-weaning feed intake and the inclusion of sensory additives upregulates the expression of immune and metabolic related genes in the small intestine, fostering improved gut health probably by facilitating the transition of feed intake during weaning. **Key Words:** Sensory additives, Weaning, Immune response

## Theme V: Mucosal Immunity and Pathogenesis and the Role of the Digestive Tract in the Maintenance of Health

**236 The intestinal barrier. Too much of a good thing?** J.R. Turner*, *Laboratory of Mucosal Barrier Pathobiology, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA.* Tissue barriers, formed by epithelial cells and intercellular tight junctions, are essential for health. For example, cellular damage and disruption of the nearly absolute epidermal barrier can lead to systemic disease and, if extensive, death. In contrast, physiological function requires selectively permeable barriers at other sites, such as the intestines and renal tubules. This poses a particular challenge for the gut, which must keep harmful luminal microbes at bay while supporting both transcellular and paracellular nutrient transport. Paracellular flux can occur via distinct pore and leak pathways, which accommodate ions and water or larger macromolecules, such as small nutrients and proteins, respectively. Available permeability probes only report leak pathway permeability, as they are too large to traverse the pore pathway. Nevertheless, extensive data indicate that pore pathway flux can improve health, promote pathogen clearance, regulate mucosal immune responses, and modify the microbiome. This presentation will focus on the cell biology, physiology, and pathobiology of intestinal pore pathway function and potential for therapeutic intervention. **Key Words:** mucosal, tight junctions, immunity

**237 Evaluating the impact of F18 Enterotoxigenic *E. coli* ileum attachment on Notch and Wnt signaling during early disease in nursery pigs.** E. M. Due*[1], K. A. Miller[1], E. R. Burrough[1], E. T. Helm[2], and N. K. Gabler[1], [1]*Iowa State University, Ames, IA, USA*, [2]*Virginia Polytechnic Institute and State University, Blacksburg, VA, USA.* The Notch and Wnt signaling pathways critically regulate intestinal homeostasis and regeneration during health and disease. Our objective was to evaluate the effect of enterotoxigenic F18 *Escherichia coli* (ETEC) attachment on intestinal function and Notch and Wnt signaling. Twenty-four FUT1 genotyped pigs (5.7 ± 0.44 kg BW) were assigned to 4 treatments (n=6) consisting of: 1) non-challenged (NC) or challenged with F18 ETEC at 2) $10^7$ cfu/mL ($10^7$), 3) $10^8$ cfu/mL ($10^8$) or 4) $10^9$ cfu/mL ($10^9$). Following two persistent days of scouring, on day post-inoculation (dpi) 5, pigs were euthanized to assess ileum F18 ETEC attachment by *in situ* hybridization, ileal histopathology, and markers of intestinal proliferation and differentiation. Data were analyzed for the main effects of treatment and challenge (NC vs. ETEC Challenge). Fecal scores were greater for all challenged pigs compared to NC ($P < 0.01$). Compared to NC, dpi 0-5 ADG tended to be lowest in the $10^8$ pigs (0.26 vs 0.10, respectively, $P = 0.07$). Fecal and ileal contents *F18* and *LT* gene abundance increased in all treatments compared to NC ($P < 0.01$). *In situ* assessment of attachment reported percentage of *F18* positive area increased with challenge dose (0.01, 0.89, 0.33, and 1.28%, respectively, $P < 0.01$). Ileum immunohistochemistry assessment of the cell proliferation marker, Ki67, tended to decrease in $10^9$ compared to NC and $10^8$ ($P = 0.08$). *Hes1* abundance was attenuated in $10^9$ compared to NC ($P < 0.05$). Compared to NC, ETEC challenge increased *SGLT1* ($P = 0.04$), tended to increase *Notch1* ($P = 0.08$) and decrease *Hes1* ($P = 0.08$), and did not change *CFTR* abundance in ileal tissue ($P > 0.10$). *Wnt4*, *BCAT* and *ATOH* mRNA did not differ ($P > 0.10$). In conclusion, F18 ETEC attachment increases with greater cfu dose. Further, ETEC may result in decoupling Hes1 from Notch signaling. Thus, modulating Hes1 could balance absorptive and secretory cell proportions in peak ETEC conditions with epithelial dysregulation. **Key Words:** ETEC, Notch, Wnt

**238 Enhancing intestinal health and antioxidant defense in weaned piglets treated with organic acids.** S. A. Flores[1], P. H. Pereira[1], I. C. Tavares[1], R. F Chaves[2], S. R. Silva Júnior[3], K. V. Z. Augusto[4], G. Heim[5], C. A. P. Garbossa[6], and V. S. Cantarelli*[1], [1]*Faculty of Animal Science and Veterinary Medicine, Federal University of Lavras, Lavras, Minas Gerais, Brazil*, [2]*AnimalNutri Ciência e Tecnologia, Patos de Minas, Minas Gerais, Brazil*, [3]*University of Minnesota, Saint Paul, Minessota, United States of America*, [4]*Trouw Nutrition, Campinas, São Paulo, Brazil*, [5]*Trouw Nutrition, Amersfoort, Netherlands*, [6]*School of Veterinary Medicine and Animal Sciences, University of São Paulo, Pirassununga, São Paulo, Brazil.* Weaning is a critical phase in piglets' lives, often causing stress and health challenges. This study examined the effects of organic acids (OA) in drinking water on the antioxidant capacity and intestinal health of nursery piglets, focusing on how

these acids influence intestinal parameters without using antibiotics as growth promoters. A total of 112 weaned barrows (average weight: 5.17 kg, ~20 days old) were assigned to a complete randomized block design. The piglets were divided into four treatments: a negative control (NC, no additives), a positive control (PC, colistin in the diet), and two OA treatments (OA1: 1.0 mL/L, OA2: 2.0 mL/L). The animals had ad libitum access to feed and water and were orally challenged with *E. coli* K88$^+$ on days 3 and 4 post-weaning. Jejunal antioxidant enzyme activity (catalase [CAT], superoxide dismutase [SOD], glutathione S-transferase [GST], and lipid peroxidation [LPO]), intestinal morphometry (villus height and crypt depth), and volatile fatty acid (VFA) concentrations in cecal contents were measured. Tissue and content samples were collected post-mortem on day 9. OA1 significantly enhanced SOD activity ($P < 0.001$) compared to all other treatments. CAT activity was higher in the PC and OA groups compared to NC, and GST activity tended to increase with OA1 ($p = 0.0364$), while LPO levels and glutathione concentrations did not differ significantly among treatments. Furthermore, OA2 showed a numerical increase in villus height (37% higher than NC) however, it did not differ statistically. Total VFA concentrations ($p = 0.0697$), acetic acid ($p = 0.0969$), and propionic acid ($p = 0.0819$) tended to be higher in OA1. These results show that organic acids in drinking water enhance jejunal antioxidant capacity and support intestinal health in weaned piglets without relying on antibiotics, offering a promising strategy for navigating the challenges of the critical post-weaning period. **Key Words:** Post-weaning, antibiotic-free, drinking water

**239 *Salmonella*-infected myeloid cells express butyrate receptors in the lower porcine intestinal tract.** S.R. Becker*[1] and C.L. Loving[2], [1]*Immunobiology Graduate Program, Iowa State University, Ames, IA, United States*, [2]*USDA-ARS-National Animal Disease Center, Ames, IA, United States*. Diet-induced production of the short-chain fatty acid butyrate can modulate intestinal and immune function and is associated with reduction of foodborne *Salmonella* shedding in pigs. The mechanism(s) of *Salmonella* reduction associated with butyrate are unclear, including the interaction of *Salmonella* and intestinal macrophages that may express butyrate receptors. Thus, we sought to determine if cecum and colon macrophages express butyrate receptors and whether *Salmonella* infects the same population of cells. Four, four-week-old pigs were oronasally inoculated with 10^8 CFU of *Salmonella* I 4,[5],12:i:-. Formalin-fixed, paraffin-embedded intestinal tissues were collected at seven days post-infection to assess butyrate receptor expression using RNAscope and *Salmonella* localization via immunohistochemistry. IBA-1 labeling was used to identify myeloid cells, including macrophages. A minimum of 8 frames were acquired per animal, per tissue for each butyrate receptor gene. Tissues from non-infected animals, as well as non-stained slides, were included as controls. Butyrate receptors *FFAR2, FFAR3,* and *HCAR2* were expressed by a fraction of IBA-1+ cells in the lower intestine. *Salmonella* was detected in IBA-1+ myeloid cells, including instances in which *Salmonella*+IBA-1+ cells expressed *FFAR2, FFAR3,* or *HCAR2.* In conclusion, we have identified porcine intestinal myeloid cells infected with *Salmonella* expressed butyrate receptors *FFAR2, FFAR3, or HCAR2,* suggesting butyrate may impact *Salmonella* infection or clearance of infected myeloid cells. Thus, diet-induced production of butyrate may alter *Salmonella* infection of intestinal myeloid cells. **Key Words:** *Salmonella,* Butyrate receptors, Intestinal myeloid cells

**240 Interrogating porcine intestinal immune status to enhance disease resilience.** C.L. Loving*[1], J.E. Wiarda[1], S. R. Becker[2], and K.A. Byrne[1], [1]*USDA-ARS-National Animal Disease Center, Ames, IA, United States*, [2]*Immunobiology Graduate Program, Iowa State University, Ames, IA, United States.* The gastrointestinal tract the largest immune organ in the body. It maintains the complex balance between reaction and regulation in serving as a barrier to the outside world, including the microbiota. As we focus on maximal growth with minimal resources and waste in pork production, interrogation of digestive physiology cannot exclude intestinal immunity. The objective of our research group is to enhance understanding of intestinal immunity for the development of approaches to enhance disease resilience. Flow cytometry is useful for identifying specific cell types but has limited capacity to deeply evaluate function. For example, age at weaning impacts the abundance of intraepithelial T cells, but it's unclear if function is also altered. Single-cell RNA-sequencing has significantly enhanced our ability to interrogate intestinal cell identity and function, and methods to identify cells in true spatial context, such as spatial transcriptomics and *in situ* hybridization, have advanced understanding even further. We have discovered unknown cell types,

including innate lymphoid cells, and identified NEUROD1$^{hi}$ enterocytes in the small intestinal villi, near epithelial T cells. Of main interest is the identification of intestinal immune cells expressing receptors for in-feed products commonly used in pig production, including b-glucan and short-chain fatty acid (SCFA) receptors. Both are expressed on intestinal macrophages and are capable of modulating macrophage function. Overall, the foundation of developing any rational approach to enhance intestinal resilience is a basic understanding of the complex interactions between the numerous cell types, and tools to interrogate cell function. **Key Words:** Intestinal immunity Single-cell RNA-sequencing

**241 Intestinal plasma cells secreting IgA regulate *Bacteroides uniformis* commensalism and are dysregulated in weaned reaction.** W.J. Tang* and H.F. Wang, *College of Animal Science, Zhejiang University, Hangzhou, Zhejiang, China.* The weaning transition in piglets is a critical period during which host-microbe mutualistic associations are established, yet the mechanisms of immune surveillance remain poorly understood. This study investigates immune-microbiota interactions during weaning and their impact on gut homeostasis. We show that weaning stress induces varying degrees of inflammation in the gut, accompanied by a shift in B cell responses toward immunoglobulin A (IgA) production and alterations in the composition of IgA-coated fecal bacteria. We performed a longitudinal analysis of the gut microbiota, which revealed a high abundance of *Bacteroides* in the gut of weaned piglets. Additionally, single-cell profiling of intestinal tissues showed that weaning stress enriched B cell-related signaling pathways within the enterocyte population. In the WP group, infiltrating B cells were primarily CD138+ plasmacytes (PCs), which shifted toward an IgA-producing cluster. Further analysis revealed that, in the low IgA-coated bacterial group, the diarrheal index was significantly correlated with the abundance of *Bacteroides*, a trend not observed in the high IgA-coated group. These findings suggest that CD138+ PCs may mediate host-gut microbiota symbiosis through IgA secretion during weaning. Bacterial isolation and colonization assays identified *Bacteroides uniformis* (B. uniformis) as a key species that induces robust IgA production by CD138+ PCs. The IgA-skewed response of PCs was essential for stable *B. uniformis* colonization in the gut. In conclusion, our findings highlight the critical role of the intestinal IgA response in fostering host-microbe symbiosis during weaning. Disruption of this immune modulation compromises intestinal homeostasis, increasing susceptibility to inflammatory disorders. These insights provide new strategies for managing weaning-induced dysbiosis in livestock. **Key Words:** Weaning piglet, *Bacteroides uniformis,* IgA-coated bacteria

**242 The influence of swine dysentery on concentration of short chain fatty acid, weight of intestinal tracts and intestinal morphology in growing pigs fed diets varying in soluble and insoluble fibers from co-products.** G.I. Lee*[1,2], K.E. Bach Knudsen[1], and M.S. Hedemann[1], [1]*Department of Animal and Veterinary Sciences, Aarhus University, Tjele, Denmark*, [2]*Department of Agricultural Science, Korea National Open University, Seoul, Republic of Korea.* Swine dysentery, *caused by Brachyspira hyodysenteriae (B. hyo),* is characterized by bloody and mucous diarrhea, and it is well-known to significantly impairs intestinal morphology. The aim of this study was to investigate whether feeding growing pigs diets varying soluble and insoluble dietary fiber (DF) could alleviate the influences of *B. hyo* infection on the weight of the gastrointestinal tract (GIT), the environment of the large intestine, and intestinal morphology. Ninety-six growing pigs (26.9 ± 2.5kg; YLD) were used for the experiment. The pigs were fed four diets formulated to differ in the level and type of DF: low fiber (LF), high fiber (HF), high soluble fiber (HS) and high insoluble fiber (HI). After two weeks of adaptation to the diets, half of the pigs were infected with *B. hyo* through oral administration. The clinical signs of dysentery, including fecal scores and *B. hyo* excretion in feces were registered. Four weeks post-inoculation, half of the pigs in each group were euthanized for assessment of GIT weight, pH of digesta, concentration of short chain fatty acids (SCFA) in digesta and evaluation of intestinal morphology. The different type and level of DF in the diets did not affect the severity of the *B. hyo* infection. The absolute but not relative GIT weight in the infected pigs was lower than in non-infected pigs ($P < 0.05$). The inclusion of soluble and insoluble DF in the diets increased the absolute and relative weight of GIT ($P < 0.05$). The crypt depth and mucin area in infected pigs were significantly greater compared to non-infected pigs ($P < 0.05$). However, the dietary treatments did not affect intestinal morphology and mucin area ($P > 0.05$), likely due to the highly negative impact of the infection. Interestingly, the concentration of SCFA in digesta of cecum and colon of the infected pigs was greater than that of the non-

infected pigs in week 6 ($P < 0.05$). **Key Words:** Swine dysentery, SCFA, Intestinal morphology

**243 The therapeutic effects of oat beta-glucans in an experimental porcine model of Crohn's disease.** Dominika Szkopek*[1], Lukasz Kopiasz[2], Jaroslaw Wolinski[1], Kinga Majchrzak-Kuligowska[3], Kamil Zaworski[1], Katarzyna Dziendzikowska[2], Katarzyna Sikorska[4], Joanna Harasym[5,6], and Joanna Gromadzka-Ostrowska[2], [1]*Laboratory of Large Animal Models, The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Instytucka Str. 3, Jablonna, Poland*, [2]*Department of Dietetics, Institute of Human Nutrition Sciences, Warsaw University of Life Sciences, Nowoursynowska Str. 159C, 02-776 Warsaw, Poland*, [3]*Department of Physiological Sciences, Institute of Veterinary Medicine, Warsaw University of Life Sciences, Nowoursynowska Str. 159, 02-776 Warsaw, Poland*, [4]*Centre for Radiobiology and Biological Dosimetry, Institute of Nuclear Chemistry and Technology, Drodna Str. 16, 03-195 Warsaw, Poland*, [5]*Department of Biotechnology and Food Analysis, Wroclaw University of Economics and Business, Komandorska Str. 118/120, 53-345 Wroclaw, Poland*, [6]*Adaptive Food Systems Accelerator-Science Centre, Wroclaw University of Economics and Business, Komandorska Str. 118/120, 53-345 Wroclaw, Poland*. The purpose of this study was to evaluate the ability of low molar mass oat β-glucan (OBG) to modulate the immune response in pigs with induced colitis, as a model for inflammatory bowel disease. OBG was obtained by alkaline water extraction. The OBG solution was prepared by mixing OBG powder with ultrapure water, obtaining a 5% solution of OBG. The experiment was conducted on 32 male piglets, which were divided into 4 groups (n=8): control, colitis, colitis with a lower OBG dose (colitis_OBG_1) and colitis with a higher OBG dose (colitis_OBG_2). After the adaptation period, colitis was induced by single rectal administration of DNBS solution (2, 4-dinitrobenzene sulfonic acid, 80 mg/kg BW). Animals in the control group were administered a 0.9% NaCl solution. The OBG solution at 37°C was administered to pigs via an intragastric cannula at 2 doses corresponding to 100 or 200 mg OBG/kg BW/day. After 21 days, the animals were sacrificed, and a section of the colon was sampled. Profile of colon intraepithelial (IETs) and lamina propria (LPTs) lymphocytes were analyzed by flow cytometry. Data were analyzed using ANOVA and Kruskal-Wallis tests, with normality checked via Shapiro-Wilk test. The induced inflammation did not significantly affect the profile of IETs and LPTs T lymphocytes ($P > 0.05$). OBG treatment decreased the percentage of cytotoxic T cells ($P < 0.05$) and increased helper T cells ($P < 0.05$), which in turn raised the CD4/CD8 cell ratio ($P < 0.05$). Moreover, OBG treatment led to a reduction in the percentage of Th17 cells ($P < 0.01$). The impact of OBG on the T-lymphocyte profile suggests an anti-inflammatory effect via the modulation of the immune response. This effect was observed in the intraepithelial layer, which is the area with the highest exposure to pathogens and is affected by inflammation that damages the intestinal barrier. These findings confirm the protective role of OBG. Study funded by National Science Centre (NCN), Poland. Grant number 2022/47/B/NZ9/03123. **Key Words:** colitis, Crohn's disease, beta-glucans