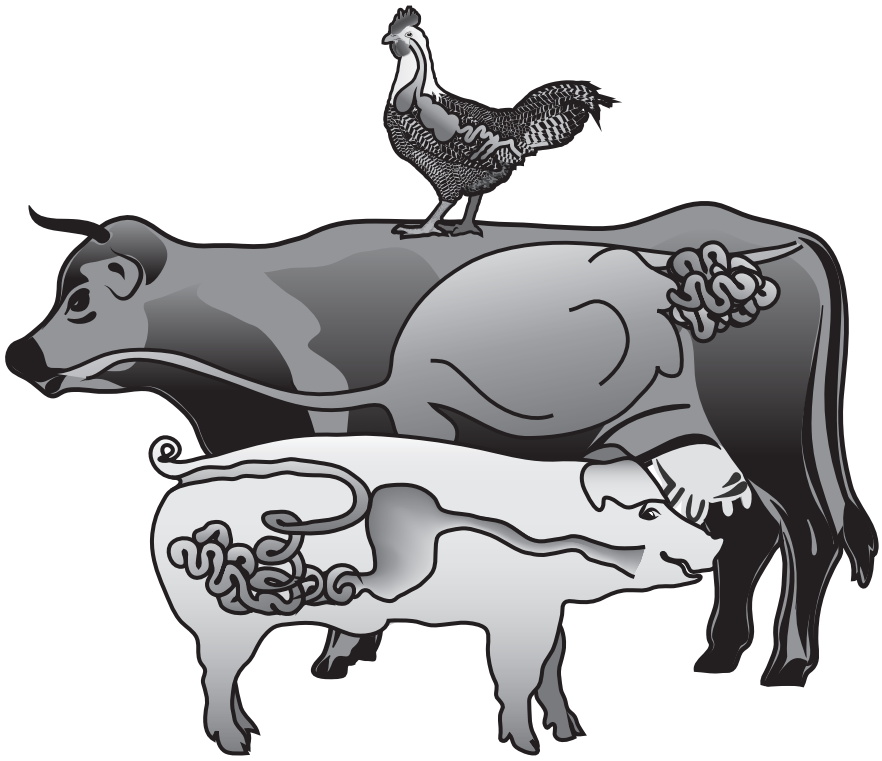
Symposium on Gut Health

in Production of Food Animals

November 6–9, 2022, St. Louis, Missouri



Program and Abstracts

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WELCOME

On behalf of the Organizing Committee for the 10th Symposium on Gut Health in Production of Food Animals, I welcome you back to St. Louis, Missouri! With the worst of the pandemic seemingly behind us, we are very happy to be back in St. Louis at an in-person Symposium.

As always, the aim of the Symposium is to bring together a group of scientists from academia, government, and industry to discuss the role of gut health in animal production and the essential role that the gut plays in establishing and maintaining animal health. The overall aim of the conference is to promote the unifying concepts that the gut drives animal health and performance. Although the gastrointestinal tract is frequently described simply as “the gut,” it is actually made up of (1) an epithelium; (2) a diverse and robust immune arm, which contains most of the immune cells in the body; and (3) the commensal bacteria, which contain more cells than are present in the entire host organism. The crosstalk between all of these interrelated components of the gut cumulatively makes the gut the basis for the well-being of animals and the motor that drives their performance. The abstracts submitted to the Symposium define the links and mechanisms that interconnect the three components of the gut and how each can be manipulated to improve animal health.

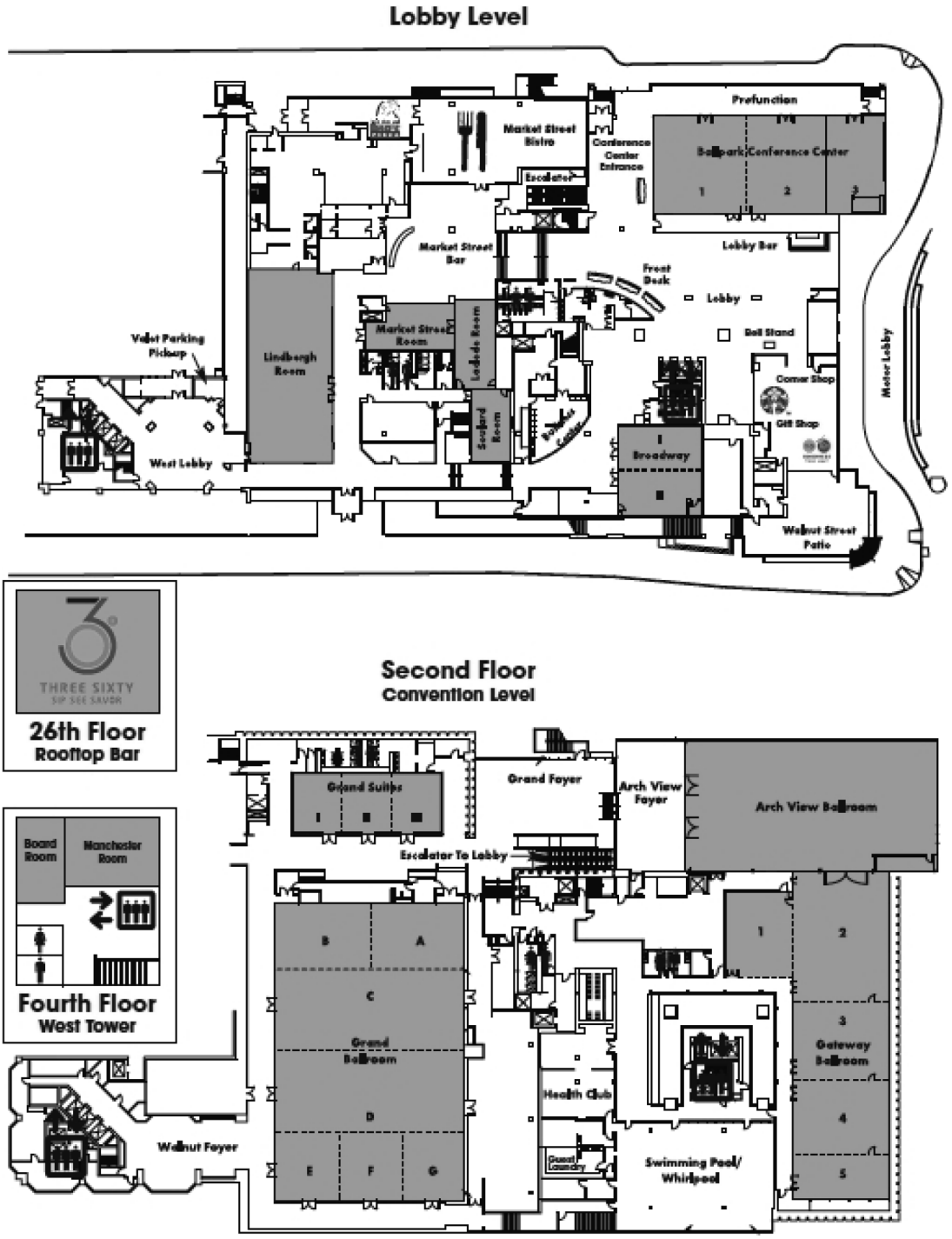
As in the past, this year we have invited three distinguished plenary speakers who will cover current research topics in avian, bovine, and porcine gut health. Please take advantage of the presence of these scientists to engage in productive talks and develop collaborations between different laboratories, to further the science of gut health.

I encourage all of you to please take advantage of the informal nature of the symposium: it was planned this way to encourage interaction between scientists. I again ask that senior researchers make a special effort to engage with the graduate students who are attending and presenting. Remember that, whatever your research specialty or food animal commodity, we are all working together to improve food quality for the consumer.

Welcome again, and enjoy the Symposium and your stay in St. Louis!

Mike Kogut
Chair, Organizing Committee

Hilton St. Louis at the Ballpark



Program

Sunday, November 6

5:00 PM - 7:00 PM Registration: Grand Foyer

Monday, November 7

7:00 AM - 8:00 AM Breakfast: Arch View Ballroom

7:00 AM - 5:00 PM Registration: Grand Foyer

SESSION 1:

Chair: Mike Kogut, USDA
Salon A, B, C

8:00 AM **Invited presentation: Getting to the guts of food animal health.**
L. Broom*, *Gut Health Consultancy, UK*.

9:00 AM **Subclinical doses of dietary fumonisins and deoxynivalenol alter the cecal microbiota of broiler chickens.**
R. Shanmugasundaram*[1], W. A. Hakeem[2], M. M. Dycus[3], T. J. Applegate[2], and J. Lourenco[3], [1]*Toxicology and Mycotoxin Research Unit, US National Poultry Research Center, Agricultural Research Service, US Department of Agriculture*, *Athens, GA, USA*, [2]*Department of Poultry Science, University of Georgia*, *Athens, GA, USA*, [3]*Department of Animal and Dairy Science, University of Georgia*, *Athens, GA, USA*.

9:30 AM **Mycotoxin deactivator promotes the intestinal health of broiler chickens challenged with *Salmonella* and mycotoxins.**
D. P. Preveraud*[1], M. Ingberman[2], B. Castello Branco Beirão[2], N. Simarro-Fagundes[3], W. Quinteiro-Filho[3], and H. M. Yakout[4], [1]*Adisseo France SAS*, *Antony, France*, [2]*Imunova Análises Biologicas*, *Curitiba, Brazil*, [3]*Adisseo Brasil*, *São Paulo, Brazil*, [4]*Adisseo North America*, *Alpharetta, GA, USA*.

10:00 AM Coffee Break: Grand Foyer
Sponsored by Jefo Nutrition Inc.

10:30 AM **Liver microbiome changes identified by transcriptome sequencing in young calves with feed-induced acidosis.**
W. Li*[1], A. Larsen[1,2], and B. Murphy[1,2], [1]*US Dairy Forage Research Center*, *Madison, WI, USA*, [2]*Oak Ridge Institute for Science and Education*, *Oak Ridge, TN, USA*.

11:00 AM **North American wild ruminants are protected from toxic dietary alkaloids by rumen-located fungi.**
S. Grace[1], J.-L. Borgogna[1], M. Elshahed[2], L. McNew[1], B. Bothner[1], C. Carr[1], and C. J. Yeoman*[1], [1]*Montana State University*, *Bozeman, MT, USA*, [2]*Oklahoma State University*, *Stillwater, OK, USA*.

11:30 AM **Chronic stress, chronic inflammation, and mitochondria disfunction: The silent killers.**
G. Tellez-Isaias*, J. D. Latorre, and B. M. Hargis, *University of Arkansas*, *Fayetteville, AR, USA*.

12:00 PM - 1:00 PM Lunch: Arch View Ballroom

Poster Session: Grand Foyer

P100 **Microbial interventions to improve gut health in neonatal calves.**
R. Nakandalage*[1,2], L. L. Guan[1], and N. Malmuthuge[2], [1]*University of Alberta*, *Edmonton, AB, Canada*, [2]*Agriculture Agri-Food Canada*, *Lethbridge, AB, Canada*.

P101 **Digital PCR as a new highly sensitive method in chicken cytokine profiling.**
G. Giovagnoni[2], F. Perry*[1], B. Anderson-Coughlin[1], K. Kniel[1], B. Tugnoli[3], A. Piva[1,3], E. Grilli[2,4], and R. Arsenault[1], [1]*Department of Animal and Food Sciences, University of Delaware, Newark*, *Newark, DE, USA*, [2]*DIMEVET, Dipartimento di Scienze Mediche Veterinarie, Università di Bologna*, *Bologna, Italy*, [3]*Vetagro S.p.A*, *Reggio Emilia, Italy*, [4]*Vetagro Inc.*, *Chicago, IL, USA*.

P102 **Citrus-based additive characterization to explain microbiota modulation of peripartum sows and beneficial consequence on performances.**
S. Cisse*[2,3], R. Cornet[1,3], E. Belz[2], M. E. A. Benarbia[2,3], O. Zemb[4], and D. Guilet[1,3], [1]*EA 921 SONAS*, *Beaucouzé, Maine et Loire, France*, [2]*Nor-Feed SAS*, *Beaucouzé, Maine et Loire, France*, [3]*Joint Lab ANR FeedInTech*, [4]*UMR GenPhySE, INRAE*, *Toulouse, Haute-Garonne, France*.

P103 **Differential growth of *Campylobacter jejuni* in mixed populations of porcine fecal, bovine fecal, or bovine ruminal microbes.**
R. C. Anderson*, M. E. Hume, N. A. Krueger, R. B. Harvey, T. L. Poole, and T. L. Crippen, *USDA, Agricultural Research Service, Southern Plains Agricultural Research Center, Food and Feed Safety Research Unit*, *College Station, TX, USA*.

P104 **Multistrain probiotics positively affect the growth performance, meat quality and alter the fecal microbiota in broiler chickens.**
C. B. Lim*, Q. Q. Zhang, and I. H. Kim, *Department of Animal Resource and Science, Dankook University*, *Cheonan, South Korea*.

P105 **Effect of Jefo P(OA+EO+PC) to control coccidiosis and reduce intestinal lesions.**
C. Roy*, L. Lahaye, H. Salgado, R. Scott-Delaunay, and E. Santin, *Jefo Nutrition Inc.*, *St-Hyacinthe, QC, Canada*.

P106 **Use of a fermented product with saponins to reduce intestinal lesions during coccidiosis challenge in broilers.**
H. H. Salgado*, L. Ludovic, C. Roy, R. Scott-Delaunay, and E. Santin, *Jefo Nutrition Inc., Saint-Hyacinthe, QC, Canada*.

P107 **Effects of supplemental encapsulated butyric acid and zinc on rumen morphometrics and small intestine histology of finishing lambs abruptly transitioned to a grain-based diet.**
F. L. Francis*[1], B. B. Grimes[1], T. L. Maia Ribeiro[1], E. R. Gubbels[1], D. Lafleur[2], J. E. Hergenreder[2], and Z. K. Smith[1], [1]*South Dakota State University*, *Brookings, SD, USA*, [2]*Kemin Industries Inc.*, *Des Moines, IA, USA*.

P108 **Broiler gastrointestinal tract responses to commercial diets containing different levels of a yeast fermentate.**
L. A. Wythe*[1], D. K. Dittoe[1], A. Scheaffer[2], and S. C. Ricke[1], [1]*Meat Science and Animal Biologics Discovery Program, Department of Animal and Dairy Science, University of Wisconsin*, *Madison, WI, USA*, [2]*Harvest Fuels*, *Walhalla, ND, USA*.

P109 **Effects of different stocking density and phytogenic feed additives dosage levels on growth performance, nutrient digestibility, blood profile, and behavior change of growing-finishing pigs.**

H. Cho*[1], M. Song[3], J. Lee[2], H. Oh[1], Y. Kim[1], S. Jang[1], J. An[1], Y. Go[1], D. Song[1], S. Cho[4], D. Kim[4], M. Kim[4], H. Kim[5], and J. Cho[1], [1]*Chungbuk National University*, *Cheongju-si, Chungcheongbuk-do, Republic of Korea*, [2]*University of Georgia*, *Athens, GA, USA*, [3]*Chungnam National University*, *Daejeon, Republic of Korea*, [4]*Eugenebio*, *Suwon-si, Gyeonggi-do, Republic of Korea*, [5]*Dankook University*, *Cheonan-si, Chungcheongnam-do, Republic of Korea*.

P110 **Phytogenic feed additives alleviate pathogenic *Escherichia coli*-induced intestinal damage through improving barrier integrity and inhibiting inflammation in weaned pigs.**
S. Chang*[1], M. Song[2], J. Lee[3], H. Oh[1], Y. Kim[1], J. An[1], Y. Go[1], D. Song[1], H. Cho[1], S. Cho[4], D. Kim[4], M. Kim[4], H. Kim[5], and J. Cho[1], [1]*Chungbuk National University*, *Cheongju-si, Chungcheongbuk-do, Republic of Korea*, [2]*Chungnam National University*, *Daejeon, Republic of Korea*, [3]*University of Georgia*, *Athens, GA, USA*, [4]*Eugenebio*, *Suwon-si, Gyeonggi-do, Republic of Korea*, [5]*Dankook University*, *Cheonan-si, Chungcheongnam-do, Republic of Korea*.

P111 **Survey of yeast populations in fermented feed across the United States.**
F. R. Mazza*, J. S. Thompson, and A. H. Smith, *Arm & Hammer Animal and Food Production*, *Waukesha, WI, USA*.

P112 **The effect of reduced whey protein and semi-moisture diet with different energy content on growth performance and nutrient digestibility in newly weaned piglets.**
C. B. Lim*, M. M. Hossain, and I. H. Kim, *Department of Animal Resource and Science, Dankook University*, *Cheonan, South Korea*.

P113 **Meta-analysis of the effect of sodium butyrate supplementation on the performance and intestinal morphology of postweaning piglets.**
L. Arnalot, W. Lambert, and T. Chalvon-Demersay*, *METEX NØØVISTAGO*, *Paris, France*.

1:30 PM - 2:30 PM Coffee Break: Grand Foyer
Sponsored by Adisseo

SESSION 2

Chair: Mike Kogut, USDA-ARS
Salon A, B, C

3:00 PM **Invited Presentation: Microencapsulated essential oils as antibiotic alternatives in broiler chickens.**
C. Yang[1], Q. Wang[2], M. S. Diarra[2], J. Gong[2], and C. Yang*[1], [1]*Department of Animal Science, University of Manitoba*, *Winnipeg, MB, Canada*, [2]*Guelph Research and Development Centre, Agriculture Agri-Food Canada*, *Guelph, ON, Canada*.

4:00 PM **A blend of botanicals protects cultured enterocytes from damages induced by an inflammatory challenge.**
A. Bonetti*[1], A. Toschi[2], A. Piva[1,2], and E. Grilli[1,3], [1]*DIMEVET, Department of Veterinary Medical Sciences, University of Bologna*, *Ozzano dell'Emilia (BO), Italy*, [2]*Vetagro S.p.A*, *Reggio Emilia, Italy*, [3]*Vetagro Inc.*, *Chicago, IL, USA*.

4:30 PM **The importance of standardized protocol for microbiota analysis.**
M. Proszkowiec-Weglarz*[1], P. M. Campos[1,2], and J. Shao[2], [1]*USDA, ARS, NEA, ABBL*, *Beltsville, MD, USA*, [2]*USDA, ARS, NEA*, *Beltsville, MD, USA*.

5:30 PM - 7:00 PM Reception: Arch View Ballroom

Tuesday, November 8

8:00 AM - 9:00 AM Breakfast: Arch View Ballroom

8:00 AM - 5:00 PM Registration: Grand Foyer

SESSION 3

Chair: Mike Kogut, USDA-ARS
Salon A, B, C

9:00 AM **Invited Presentation: Gastrointestinal adaptation: Rapid functional and retrogressive changes but slow proliferation.**
G. Penner*, *Department of Animal and Poultry Science, University of Saskatchewan*, *Saskatoon, SK, Canada*.

10:00 AM **Broiler gut barrier function enhancement by a synergistic in-feed technology under dietary-induced chronic inflammation in real farming conditions.**
A. Khadem*[1,2], E. Griela[3], M. Sevastiyanova[1], K. C. Mountzouris[3], and C. Gougoulias[1], [1]*Innovad® NV/SA*, *Berchem, Belgium*, [2]*Laboratory of Animal Nutrition, Faculty of Veterinary Medicine, Ghent University*, *Heidestraat 19, 9820 Merelbeke, Belgium*, [3]*Laboratory of Nutritional Physiology and Feeding, Department of Animal Science, Agricultural University of Athens*, *Athens, Greece*.

10:30 AM Coffee Break: Grand Foyer
Sponsored by Optum Immunity

11:00 AM **Single-cell RNA sequencing (ScRNA seq) technology, a powerful tool to screen new solutions with the potential to impact NetB and *Clostridium perfringens* cytotoxicity.**
M. Bernardeau*[1,4], M. A. Brennan[2], G. Saxer[2], A. Z. Rosenthal[2,3], K. Gibbs[1], and J. P. Meisch[2], [1]*IFF Danisco Animal Nutrition and Health*, *Oegstgeest, the Netherlands*, [2]*IFF Health and Biosciences*, *Wilmington, DE, USA*, [3]*Department of Microbiology and Immunology UNC Medical School*, *Chapel Hill, NC, USA*, [4]*Normandy University UNICAEN ABTE*, *Caen, France*.

11:30 AM **The benefits of a direct-fed microbial combination in reducing the severity of necrotic enteritis in broilers: A combined analysis.**
S. A. S. van der Klein*[1], R. K. Selvaraj[2], A. Yitbarek[1,3], and K. Gibbs[1], [1]*Danisco Animal Nutrition & Health, IFF*, *Oegstgeest, The Netherlands*, [2]*Department of Poultry Science, University of Georgia*, *Athens, GA 30602, USA*, [3]*Department of Animal Science, McGill University*, *Montreal, QC, Canada*.

12:00 PM **A blend of botanicals reduces LPS-induced inflammation and disruptive effects on apical-out porcine enteroids.**
F. Ghiselli*[1], Y. Li[2], L.-e. Yu[2], A. Piva[1,3], and E. Grilli[1,4], [1]*DIMEVET - University of Bologna*, *Ozzano dell'Emilia, Bologna, Italy*, [2]*Department of Animal and Food Sciences, University of Delaware*, *Newark, DE, USA*, [3]*Vetagro S.p.A*, *Reggio Emilia, Italy*, [4]*Vetagro Inc.*, *Chicago, IL, USA*.

12:30 PM - 2:00 PM Lunch: Arch View Ballroom
Sponsored by Jefo Nutrition Inc.

SESSION 4

Chair: Mike Kogut, USDA-ARS
Salon A, B, C

2:00 PM **Tannins in poultry production.**
M. E. Fernandez Miyakawa*[1,2], [1]*INTA*, *Hurlingham, Buenos Aires, Argentina*, [2]*CONICET*, *Buenos Aires, Argentina*.

3:00 PM **Epigenetic regulation of host defense peptide synthesis.**
M. A. Whitmore and G. Zhang*, *Oklahoma State University*, *Stillwater, OK, USA*.

4:00 PM **Software for microbiome and untargeted metabolomics in biomarker discovery.**
B. Shannon*, *BioRankings*, *St. Louis, MO, USA*.

4:30 PM - 6:00 PM Reception: Arch View Ballroom

Wednesday, November 9

8:00 AM - 9:00 AM Breakfast: Arch View Ballroom

8:00 AM - 11:00 AM Registration: Grand Foyer

SESSION 5

Chair: Mike Kogut, USDA-ARS
Salon A, B, C

9:00 AM **Comparative efficacy of different natural products based on liquid Quillaja extract administered in the feed for the control of necrotic enteritis in chickens.**
S. Decap*[1], V. Tapia[1], R. Navarro[1], and B. Lumpkins[2], [1]*Plantae Labs SpA*, *Santiago, Chile*, [2]*Southern Poultry Research*, *Athens, GA, USA*.

9:30 AM **Describing the cecal microbiota development in broilers when fed varying levels of a yeast culture product.**
L. A. Wythe*[1], D. K. Dittoe[1], A. Scheaffer[2], and S. C. Ricke[1], [1]*Meat Science and Animal Biologics Discovery Program, Department of Animal and Dairy Science, University of Wisconsin*, *Madison, WI, USA*, [2]*Harvest Fuels*, *Walhalla, ND, USA*.

10:00 AM **Types and dietary levels of resistant starches modulated growth performance and intestinal environment in broiler chickens.**
I. W. Oluseyifunmi* and O. A. Olukosi, *University of Georgia*, *Athens, GA, USA*.

10:30 AM **Rumen microbial cluster identification and its influence on rumen metabolites and growth performance of young goats.**
D. Wang[1], G. Tang[1], L. Wang[1,2], X. Chen[2], J. Yao[1], and Y. Cao*[1,2], [1]*Northwest A&F University*, *Yangling, Shaanxi, China*, [2]*Harvard Medical School*, *Boston, MA, USA*.

Session 1

100 Getting to the guts of food animal health. L. Broom*, *Gut Health Consultancy, UK.*

Studies with germ-free animals have clearly demonstrated the importance of microorganisms for proper development of a fully functional immune system and to influence an array of host physiological processes. Similarly, work with gnotobiotic animals shows that microbial colonization recapitulates host immune and physiological features, the degree to which is often dependent on the specific microbe(s), or the complexity of the microbial consortia administered. Therefore, the importance of microbial exposure for host development has been well established. Generally, the host is considered to be initially exposed to commensal microbes during the birthing or hatching process. However, recent data propose the possibility of *in-utero* or *in-ovo* colonization of the host and intestine, making this a hotly debated topic. Precisely when and what the host tissues are exposed to will likely have fundamental implications for host and immune system development, as well as potentially guiding the appropriate application of exogenous interventions seeking to influence early microbial colonization and developmental trajectory. Typically, and depending on the production system, these initial microbes will largely be of maternal or (immediate) environmental origin, acquired in a largely random process, thus seeding each individual with a unique microbiota composition, although functional profiles tend to be more similar. Together, the host, diet, and microbiome largely create biogeographical niches (e.g., substrates and O_2 availability, pH) within the gastrointestinal tract that support particular microbial communities, which challenges the collection of appropriate samples when seeking to explore these populations and their relationship with host health and growth performance. There remain many unknowns with regards to how the gut microbiome and host interact, and how these interactions manifest as relatively better or worse health or performance outcomes. We will explore important aspects of host-microbiome interactions to hopefully inspire insightful thoughts, discussion, and to encourage colleagues in their endeavors.

Key Words: Gut health, immune system, microbiota, microbiome, performance

101 Subclinical doses of dietary fumonisins and deoxynivalenol alter the cecal microbiota of broiler chickens. R. Shanmugasundaram*[1], W. A. Hakeem[2], M. M. Dycus[3], T. J. Applegate[2], and J. Lourenco[3], *[1]Toxicology and Mycotoxin Research Unit, U.S. National Poultry Research Center, Agricultural Research Service, U.S. Department of Agriculture, Athens, GA, USA, [2]Department of Poultry Science, University of Georgia, Athens, GA, USA, [3]Department of Animal and Dairy Science, University of Georgia, Athens, GA, USA.*

Fusarium toxins are one of the most common contaminants in poultry diets. Co-occurrence of fumonisins (FUM) and deoxynivalenol (DON), even at a subclinical dose, negatively affects the production performance and intestinal integrity in broiler chickens. Loss of gut integrity can be expected to alter the intestinal microbiota composition. This study evaluated the effects of subclinical doses of combined mycotoxins on the cecal microbiota composition and diversity in broiler chickens. Broiler chickens were raised on 2 diets: starter (d 0–21), and grower diet (d 22–35). A total of 240 one-day-old chicks were randomly assigned to 2 treatments, each replicated in 8 pens with 15 birds per pen. The experimental treatments were control diet and 3 mg/kg FUM + 4 mg/kg DON contaminated diet. On d35, 3 mg/kg FUM + 4 mg/kg DON contaminated diet tended to decrease the body weight by 84 g compared with the control group ($P = 0.06$). Cecal contents were collected on d 21, 28, and 35. The bacterial compositions of the cecal contents were analyzed by Illumina MiSeq sequencing of the V3–V4 region of the 16S rRNA gene. Overall, microbial richness and diversity increased ($P < 0.05$) during the studied period (d 21–35). Cecal contents of birds in the 3 mg/kg FUM + 4 mg/kg DON group had greater ($P < 0.05$) microbial evenness and diversity (Shannon index), compared with the control group. Chronic exposure of 3 mg/kg FUM + 4 mg/kg DON decreased ($P = 0.001$) the relative abundance of Proteobacteria in the cecal content, compared with the control group. At the genus level, the cecal content of birds in the 3 mg/kg FUM + 4 mg/kg DON showed decreased ($P < 0.05$) relative abundances of Acinetobacter and Pseudomonas, and a tendency ($P = 0.08$) for a lower abundance of Thermincola compared with the control group. The present findings showed that dietary FUM and DON contamination, even at subclinical levels, altered the bacterial microbiota composition and diversity in the cecal content of the birds.

Key Words: Fumonisins, deoxynivalenol, mycotoxins, 16s sequencing, cecal microbiota

102 Mycotoxin deactivator promotes the intestinal health of broiler chickens challenged with *Salmonella* and mycotoxins. D. P. Preveraud*[1], M. Ingberman[2], B. Castello Branco Beirão[2], N. Simarro-Fagundes[3], W. Quinteiro-Filho[3], and H. M. Yakout[4], *[1]Adisseo France SAS, Antony, France, [2]Imunova Análises Biologicas, Curitiba, Brazil, [3]Adisseo Brasil, São Paulo, Brazil, [4]Adisseo North America, Alpharetta, GA, USA.*

The objective of the study is to evaluate the effect on intestinal and health parameters of a mycotoxin challenge with the presence of *Salmonella* infection in broilers. For that, 230 one-day-old Ross broilers were acquired from a commercial hatchery. The animals were placed in isolators and randomly assigned to 5 different experimental groups of 46 broilers each. The treatments consist of a control group (CTRL), a mycotoxin group (MC), a mycotoxin group receiving a mycotoxin deactivator, a co-infection mycotoxin-*Salmonella* group (MSC), and a mycotoxin-*Salmonella* group receiving a mycotoxin deactivator (MSUP). On the first experimental day, the animals in MC, mycotoxin deactivator, MSC and MSUP groups started to receive the mycotoxins in their diet (mainly fumonisin, deoxynivalenol, and T-2 toxin). On the fourth experimental day, each animal of MSC and MSUP groups were challenged with 1×10^8 cfu of *Salmonella* Heidelberg by oral delivery. Then, between d 4 and 28, intestinal permeability was evaluated by oral administration of FITC-dextran, ceca content was collected to assess *Salmonella* spp. counts by most probable number and microbiota S16 sequencing, in cecal tonsil, the cytokines gene expression was measured, histomorphology was assessed from jejunum and cecum tissues, liver samples were also collected for lipoperoxidation and histopathology evaluation, and finally anti-*Salmonella* IgA was quantified in feces. Statistical analysis was performed by 2-way ANOVA with Tukey's post-hoc test ($P < 0.05$). Mycotoxin presence in the feed can significantly increase intestinal permeability, contributes to higher *Salmonella* count in ceca and anti-*Salmonella* IgA in feces, modulates cytokines gene expression, and impaired histomorphological parameters such as intestinal villus height, epithelial height, and crypt depth. This study shows that a moderate mycotoxin challenge can highly negatively affect the health of the animals and can be a prevalence factor for *Salmonella* infection as well. An antimycotoxin solution with complementary modes of action can protect the birds and promote markers of intestinal health.

103 Liver microbiome changes identified by transcriptome sequencing in young calves with feed induced acidosis. W. Li*[1], A. Larsen[2,1], and B. Murphy[2,1], *[1]US Dairy Forage Research Center, Madison, WI, USA, [2]Oak Ridge Institute for Science and Education, Oak Ridge, TN, USA.*

Liver abscesses (LAs) are ancillary to ruminal acidosis and can cause significant economic loss as the result of liver function condemnation and decreased growth and production. Currently, there are no effect diagnostic tools for early detection or prevention of LAs. Using our established model of ruminal acidosis in young calves, we investigated the effect of ruminal acidosis on the microbial community in the liver. A group of 8 calves were randomly assigned to an acidosis-inducing diet (AC) or blunting diet, with 4 animals per treatment. Rumen epithelium and liver tissues were collected at 17 wk of age right after slaughter. Total RNAs were extracted and followed by whole transcriptome sequencing. Calves fed AC showed significantly less weight gain over the course of the experiment, substantially lower ruminal pH, and significant rumen degradation comparison to the control group ($P < 0.05$). Microbial RNA reads were enriched bioinformatically and used for microbial taxonomy classification using Kraken2, and transcript abundance analysis. In the liver, a total of 29 genera showed more than 2-fold change in abundance between the treatments. Among these, *Fibrobacter*, *Treponema*, *Lactobacillus*, and *Olsenella* have been reported in abscessed liver in cattle. Additionally, 9 of these genera also significantly increased in the rumen epithelium ($P < 0.05$, fold change >2) in the AC group. In the liver, genes involved in pyruvate metabolic process, proton-acceptors and lipid metabolism pathways had significant association with the microbial community changes. Our study sheds light into host liver community changes in postweaning calves with prolonged acidosis. The concurrent shifts in microbial genus abundance in both the liver and rumen indicate potential crosstalk between the liver and rumen epithelial microbial communities. Given the important role of the liver as a metabolic organ, our findings warrant further investigation into the role of the liver microbiome in the progression of ruminal acidosis and the specific molecular events that facilitate the interactions between the gut and the liver.

Key Words: ruminal acidosis, liver microbiome, young calves

104 North American wild ruminants are protected from toxic dietary alkaloids by rumen-located fungi. S. Grace[1], J.-L. Borgogna[1], M. Elshahed[2], L. McNew[1], B. Bothner[1], C. Carr[1], and C. J. Yeoman*[1], *[1]Montana State University, Bozeman, MT, USA, [2]Oklahoma State University, Stillwater, OK, USA.*

Consumption of the toxic alkaloid, methyllycaconitine (MLA) found in tall larkspur, causes an estimated loss of 5 to 15% of Northwestern rangeland cattle annually; however, there is no evidence that wild ruminant species are similarly affected. We therefore hypothesized that the gastrointestinal microbiota of wild ruminant species might mediate protection against MLA toxicity by degrading the MLA. To examine this, 159 rumen samples were collected from pronghorn, mule and white-tailed deer, elk, moose, bighorn sheep, mountain goat, and bison harvested throughout Montana in 2019 and 2020 by volunteer hunters and assayed for total alkaloid- and MLA-specific degradation activities *in vitro*. Changes in total alkaloids were measured spectrophotometrically, and changes in MLA were measured by HPLC-MS. The results varied

by individual animal, rather than sex, age, species, or the location where animals were harvested, with 90% of specimens exhibiting between 2.1 and 70.8% degradation of total *Delphinium* species alkaloids, and all of these samples exhibiting between 0.6 and 80.8% degradation of total MLA. A separate series of experiments were performed after treating specimens with an antibiotic cocktail or after autoclave sterilization. These findings revealed that abiotic and bacterial activities made only minor contributions to total alkaloid and MLA degradation. We then performed the same experiments using 15 fungal isolates from wild rumen samples and found that 6 strains were able to degrade between 33 and 71% of total MLA. These findings suggest rumen fungi protect wild ruminants from toxic alkaloids and hypothesize they may be exploited to protect range cattle.

Key Words: rumen, fungi, toxin degradation, Larkspur toxicosis, methyllycaconitine

105 Chronic stress, chronic inflammation, and mitochondria disfunction the silent killers. G. Tellez-Isaias*, J. D. Latorre, and B. M. Hargis, *University of Arkansas, Fayetteville, AR, USA.*

Mitochondria are the "powerhouses" of cells, providing the energy and many molecular building blocks needed for metazoans to remain alive. They derive from bacteria that took up residence inside a host cell 2 billion years ago in a symbiotic relationship that altered the course of life. Mitochondria are also involved in various other metabolic processes, including signaling through mitochondrial reactive oxygen species, regulation of the membrane potential, apoptosis-programmed cell death, calcium signaling, regulation of cellular metabolism, heme synthesis reactions, steroid synthesis, and hormonal signaling, among others. Diet ingredients and the balance of the microbiome play a direct role in mitochondria biology. As a result, mitochondrial damage and subsequent malfunction are significant contributing factors to various diseases due to their influence on cellular metabolism. Stress and inflammation are innate responses in living organisms that involve hormones, immune cells, and molecular mediators. They are critical mechanisms for the survival and healing of all life forms, and various stimuli trigger them. However, the energy and vitality of healthy mitochondria are required for all the systems to function correctly during stress responses. In chronic stress and chronic inflammation, an increase in the generation of reactive oxygen species causes peroxidation of lipids in cell membranes and mitochondrial membranes, compromising cell homeostasis. This presentation will discuss the interactions between diet ingredients, gut microbiome, nervous system, immune system, endocrine system, and mitochondria during health and metabolic and gastrointestinal disorders, cancer, autoimmune diseases, myopathies, cardiovascular, and even neurological diseases, recognizing chronic oxidative stress, chronic inflammation, and mitochondrial dysfunction as the silent killers.

Key Words: mitochondria, chronic stress, chronic inflammation, microbiota, diet ingredients

Session 2

106 Microencapsulated essential oils as antibiotic alternatives in broiler chickens. C. Yang[1], Q. Wang[2], M. S. Diarra[2], J. Gong[2], and C. Yang*[1], *[1]Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada, [2]Guelph Research and Development Centre, Agriculture Agri-Food Canada, Guelph, Ontario, Canada.*

Antimicrobial resistance (AMR) has become a serious problem in poultry farms that can threaten both poultry and human health. An increasing number of studies have been conducted on exploring antimicrobial alternative such as essential oils (EO). The objective of this study was to evaluate the effects of encapsulated EO including cinnamaldehyde (CIN) and citral (CIT) alone or in combination (CIN+CIT) on growth performance, meat quality, gut health, and AMR phenotypes, genotypes, and virulence of broiler chicken fecal *Escherichia coli* isolates, and zoonosis of poultry-isolated AMR extraintestinal pathogenic *E. coli* (ExPEC). Results showed the feed efficiency ratio (FCR), mortality (%), gut lesion scores were all reduced by bacitracin and encapsulated CIN, CIT, and CIN+CIT. Cecal microbiota was modulated in birds fed bacitracin, CIN, CIT, and CIN+CIT compared with birds fed basal diets. Furthermore, vaccinated birds showed altered cecal microbiota, reduced mortality (%) but higher gut lesions compared with nonvaccinated birds. The AMR levels (%) of chicken fecal *E. coli* to most tested antimicrobials were lower in birds fed encapsulated CIN or CIN+CIT which also showed reduced prevalence (%) of some antimicrobial resistance genes and plasmids. Additionally, bird age is also a factor affecting AMR phenotypes, genotypes, and virulence of chicken fecal *E. coli*. Encapsulated CIN improved apparent ileal nutrient digestibility, intestinal duodenal and jejunal villus/crypt ratio, jejunal gene expressions for nutrient transporters, and changed cecal and ileal intestinal microbiota. The ExPEC isolated from poultry meat and feces had significant effects on reducing survival (%) of *Caenorhabditis elegans* but the relationships between antimicrobial susceptibility or number of virulence genes with pathogenicity of *E. coli* isolates were not conclusive. In conclusion, encapsulated CIN has the potential to improve growth performance, gut health, meat quality, modulating ileal and cecal microbiota, and reduce resistance level (%) to antimicrobials and prevalence of antimicrobial resistance genes and plasmids in chicken fecal *E. coli* isolates. Additionally, ExPEC isolated from poultry meat or feces may possess zoonotic potential to cause human infections.

107 A blend of botanicals protects cultured enterocytes from damages induced by an inflammatory challenge. A. Bonetti*[1], A. Toschi[2], A. Piva[1,2], and E. Grilli[1,3], *[1]DIMEVET, Department of Veterinary Medical Sciences, University of Bologna, Ozzano dell'Emilia (BO), Italy, [2]Vetagro S.p.A., Reggio Emilia, Italy, [3]Vetagro Inc., Chicago, IL, USA.*

Botanicals represent a wide class of complex compounds that naturally contain active principles with numerous biological functions. Among them, the anti-inflammatory and antioxidant actions are of key interest to support intestinal health during stressful phases. The aim of this study was to investigate the ability of a thymol-based blend of botanicals (BOT) to protect intestinal Caco-2 cells from the damages induced by an inflammatory challenge in vitro. Caco-2 cells were differentiated on porous inserts in basal medium, then treated with (CTR+) or without (CTR-) LPS+cytokines challenge, or LPS+cytokines challenge and BOT (BOT+). Transepithelial electrical resistance was daily measured for 7 d and, at the end, cells were harvested for gene expression analysis. Cells were also cultured on 96 well plates or glass coverslips, and treated for 24 h as previously described, to measure reactive oxygen species and perform immunofluorescence staining. The BOT treatment was effective in protecting enterocytes from inflammatory damages because transepithelial electrical resistance was significantly ameliorated in BOT+ group compared with CTR+ ($P < 0.05$). This outcome was supported by the increased levels of ZO-1, ZO-2, CLDN-1, CLDN-3, and OCCL observed when BOT was added to challenged Caco-2 cells. Moreover, immunofluorescence staining showed improved ZO-1 localization in BOT-treated cells. The BOT mechanism of action was related to a significant reduction in Caco-2 inflammatory response, as demonstrated by the decreased IL-6, IL-8 ($P < 0.05$), and TNFα ($P < 0.1$) levels. Furthermore, BOT improved the oxidative status of challenged enterocytes by significantly scavenging cellular reactive oxygen species. This effect was mediated by BOT ability to enhance the expression of antioxidant enzymes such as GPX-1 ($P < 0.05$) and numerically increase the mRNA levels of SOD. BOT was able to maintain epithelial integrity in vitro by helping enterocytes to face oxidative stress and inflammatory stimuli. BOT has the potential to be further investigated in vivo as a nutritional supplement to support animals' transition through stressful phases.

Key Words: botanicals, inflammation, oxidative stress, intestinal integrity

108 The importance of standardized protocol for microbiota analysis. M. Proszkowiec-Weglarz*[1], P. M. Campos[1,2], and J. Shao[2], *[1]USDA, ARS, NEA, ABBL, Beltsville, MD, USA, [2]USDA, ARS, NEA, Beltsville, MD, USA.*

Bacteria, the major component of chicken gastrointestinal tract (GIT) microbiota, play an important role in health, nutrition, host physiology regulation, GIT development, and growth. Recently, the microbial community profiling method based on the 16S rRNA sequencing approach has become the most popular to determine the taxonomic composition and diversity of chicken microbiota. The 16S rRNA profiling consists of many steps such as: sample collections and storage, DNA isolation, 16S primer selections, 16S rRNA PCRs, libraries preparations and indexing, sequencing, raw data analysis (pipeline or software selection), operational taxonomic unit/amplicon sequence variant picking, taxonomic database selection, diversity analysis, and statistical analysis. Currently, there is no standardized protocol for 16S determination in chicken samples. It has been shown that primer design, library preparation, DNA isolation methods, and PCR amplification artifacts can introduce unique biases that can affect community structure, richness, and microbial population analysis and lead to over- or under-representation of individual bacteria within communities. Moreover, different sequencing platforms and bioinformatics pipelines can affect the average relative abundance of microbiota and shape the taxonomic community profiles. Although the experiments in chicken microbiota studies are commonly standardized and based on identical breeds, the results are often contradictory and depend on the used animal (breed, age, sex), the experimental design (feeding and sampling), and DNA extraction and sequencing methods. Our own data indicate that the choice of 16S primers as well as taxonomic database selection has significant effects on microbiota analysis and data interpretation. Therefore, it is difficult to compare data and correlate results originating from a different protocol. The development of a standardized protocol for microbiota profiling in chickens, similar to the one used in human microbiota research, is needed to obtain comparable data sets for poultry microbiota.

Key Words: Chickens, methodology, 16S

Session 3

109 Gastrointestinal adaptation: Rapid functional and retrogressive changes but slow proliferation. G. Penner*, *Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.*

The gastrointestinal tract (GIT) is a key organ system facilitating, inter alia, movement of digesta, digestion, nutrient absorption, host-microbial communication, and must regulate permeability to control immunogenic exposure systemically. As a large consumer of maintenance energy, disruptions in nutrient supply can cause marked changes in GIT size and function. Although data on reticulo-ruminal responses are abundant, less information is available regarding postruminal regions. That said, greater nutrient supply increases the proliferative responses of the reticulo-rumen and intestinal regions. However, the increases in size and expansion of surface area are relatively slow suggesting these are longer-term adaptive responses. To compensate, cell activity, at least in the reticulo-rumen, increases rapidly to facilitate greater nutrient transport. On the other hand, feed deprivation, and periods of low feed intake markedly reduce absorptive surface area. Interestingly, the reduction in absorptive surface area through retrogressive adaptational responses is very rapid with greater than 50% reductions occurring within 5 d of low feed intake. Additionally, exposure to low feed intake decreases the rates of SCFA absorption and transiently increases paracellular permeability. Used as a common challenge model, ruminal acidosis also causes alterations throughout the GIT with reduced rates of short-chain fatty acid transport and altered permeability. Although paracellular permeability recovers rapidly following a challenge (low feed intake or ruminal acidosis), recovery responses for nutrient transport occur gradually, likely related to the gradual recovery for dry matter intake. The basal diet fed may also alter these responses as diets high in long-chain polyunsaturated fatty acids may decrease passive apical uptake of propionate and butyrate while increasing risk for paracellular permeability. Thus, the gastrointestinal tract epithelium adapts to modulate nutrient absorption and paracellular permeability using both short-term and long-term mechanisms.

110 Broiler gut barrier function enhancement by a synergistic in-feed technology under dietary-induced chronic inflammation in real farming conditions. A. Khadem*[1,2], E. Griela[3], M. Sevastiyanova[1], K. C. Mountzouris[3], and C. Gougoulias[1], *[1]Innovad® NV/SA, Berchem, Belgium, [2]Lab of Nutrition, Faculty of Veterinary Medicine, Ghent University, Heidestraat 19, 9820 Merelbeke, Belgium, [3]Laboratory of Nutritional Physiology and Feeding, Department of Animal Science, Agricultural University of Athens, Athens, Greece.*

Dysfunction of the intestinal barrier has been associated not only with impaired nutrient absorption and reduced growth performance in broilers but also with increased microbial translocation and disease risk. Here, we evaluated the protective gut barrier effects of 1 or 2 kg/ton feed of Lumance® (Innovad, Belgium; esterified butyrate, combined with plant extracts, essential oils, and other fatty acids) in a dietary-induced chronic inflammation model (high NSP diet: 60% Wheat + 5% rye without NSPase and coccidiostats), via the distal-jejunal mRNA gene expression of mucins and tight-junction proteins in broilers. Importantly, the pens (n = 8 pens/treatment; n = 30 birds/pen) were housed inside a commercial production unit of 55,000 broilers so that the experimental birds could get exposed to the same (real) farming conditions. One broiler per pen was randomly selected on d 28 and 35 to determine the intestinal expression of tight junctions and the plasma level of FITC-dextran (the latter only at d 28). Analysis of variance with Tukey post-hoc analysis revealed that Lumance® 1 and 2 kg/ton, when compared with the control treatment, significantly increased the BW of broilers at d 35 by 3 and 4%, respectively ($P = 0.040$) and reduced the FCR (1.64, 1.62 and 1.57, respectively; $P = 0.01$). Lumance® 1 and 2 kg/ton resulted in a profound increase in intestinal expression of *Claudin-1*, *Occludin*, and *Mucin-2* (approximately, a 4- and 5-fold increase for the former and a 2- and 3-fold increase, respectively, for the latter 2 genes) compared with the control, both at d 28 and 35 ($P < 0.005$ in all cases). Additionally, a trend in reduction of FITC-d levels in plasma ($P = 0.098$) was seen at d 28 in birds receiving Lumance® compared with the control. In this study, broilers under chronic inflammation in real conditions, when fed with an in-feed synergistic technology conferred a superior protective effect on gut barrier function, accompanied by enhanced growth performance. The positive results warrant further work for the elucidation of related mechanisms.

Key Words: Intestinal barrier function, gene expression, tight junctions, growth performance, FITC-dextran

111 Single cell RNA sequencing (ScRNA seq) technology, a powerful tool to screen new solutions with the potential to impact NetB and *Clostridium perfringens* cytotoxicity. M. Bernardeau*[1,4], M. A. Brennan[2], G. Saxer[2], A. Z. Rosenthal[2,3], K. Gibbs[1], and J. P. Meisch[2], *[1]IFF Danisco Animal Nutrition and Health, Oegstgeest, the Netherlands, [2]IFF Health and Biosciences, Wilmington, DE, USA, [3]Department of Microbiology and Immunology UNC Medical School, Chapel Hill, NC, USA, [4]Normandy University UNICAEN ABTE, Caen, France.*

Necrotic enteritis (NE) caused by *Clostridium perfringens* is a reemerging threat to the poultry industry following mounting pressure to reduce antibiotic use. Alternatives to antibiotics are known for apparent inconsistent effects. As a means for improving consistency to develop next generation solutions, the etiology and characteristics of gut health challenges must be fully elucidated. Here we show the power of scRNaseq technology to analyze and decipher the effect of secreted metabolites produced by 2 direct-fed microbials (DFM) *Lactobacillus acidophilus* and the *Bifidobacterium animalis* strain, on the production of NetB (the pore-forming toxin considered as the main virulence factor among a variety of different toxins) and the cytotoxicity of a pathogenic (or NE inducing) *C. perfringens* strain. The combined secretomes of both strains reduced production of NetB and overall cytotoxicity of *C. perfringens*. Treating HT-29 cells with supernatant from *C. perfringens* grown in the presence of both DFM secretomes reduced *C. perfringens* cytotoxicity by 60% (P-value < 0.05). To better understand the effect of the DFM secretomes on the biology of *C. perfringens* and regulation of pathogenicity, a novel scRNaseq method developed internally, was used to investigate the transcriptome of single cells after growth in the presence and absence of the DFM secretome. We report, for the first time, heterogeneous gene expression in *C. perfringens* populations, with the *netB* toxin gene being primarily expressed by a subset of cells. We further show that the *B. animalis* secretome changed overall gene expression and community organization of *C. perfringens* population with an 8-fold reduction in *netB* expression (P-value < 6×10^{-7}). These *in vitro* results introduce a novel strategy to identify new targets within a population of *C. perfringens.* Consequently, this technology and information released thereof can be applied for the development of antibiotic alternatives and can guide recommendations for use in production systems.

Key Words: poultry, necrotic enteritis, NetB, scRNAseq, virulence

112 The benefits of a direct-fed microbials combination in reducing the severity of necrotic enteritis in broilers: A combined analysis. S. A. S. van der Klein*[1], R. K. Selvaraj[2], A. Yitbarek[1,3], and K. Gibbs[1], *[1]Danisco Animal Nutrition & Health, IFF, Oegstgeest, The Netherlands, [2]Department of Poultry Science, University of Georgia, Athens, GA, USA, [3]Department of Animal Science, McGill University, Montreal, Quebec, Canada.*

Necrotic enteritis (NE) onset in broilers is complex, suggesting successful nonantibiotic intervention strategies should be multipronged. This combined analysis of 2 trials evaluated the effects of a direct-fed microbials combination on gut integrity in NE-induced broilers. Treatments consisted of an unchallenged control (UC), an *Eimeria maxima* and *Clostridium perfringens* challenge control (CC), and CC supplemented with a direct-fed microbials blend (DFM). Live performance was recorded up to d 28. NE lesion scoring and fecal oocyst count (OPG) was measured at d 21. Jejunum Claudin-1 (Cl1), and Claudin-2 (Cl2) mRNA expression was analyzed on d 21 and 28. Gut permeability was evaluated in one study using a FITC-assay at d 28. Combined data were analyzed using a mixed model with trial as a random effect and treatment as a fixed effect. Compared with the CC, the DFM treatment improved body weight by 11% (P = 0.077) and FCR by 10% (P < 0.001), and reduced NE-related mortality from 12 to 1% (P < 0.001). Indicative of the mild NE model, average lesions scores increased from 0 to 1.25 in the CC compared with the UC. The DFM treatment reduced the average lesion score to 0.48 (P < 0.001). The DFM treatment also reduced oocyst shedding by 74% compared with the CC and showed a 2.6-fold increase in Cl1 expression at d 21 (P = 0.040). At d 28, the DFM treatment improved gut permeability by 50% (P = 0.012) and increased Cl1 expression 2.3-fold (P = 0.003). These results indicate that the DFM combination significantly enhanced gut integrity, reduced gut permeability, and reduced OPG cycling during a NE challenge. The study demonstrates the added value of a DFM, but additionally creates understanding of the origin of the efficacy. Analyzing and evaluating multiple parameters, including performance, host characteristics, disease indicators, and their development over time, leads to a more holistic understanding of gut health. This allows for further successful development of antibiotic alternatives.

Key Words: poultry, DFM, necrotic enteritis, FITC, tight junctions

113 A blend of botanicals reduces LPS-induced inflammation and disruptive effects on apical-out porcine enteroids. F. Ghiselli*[1], Y. Li[2], L.-e. Yu[2], A. Piva[1,3], and E. Grilli[1,4], *[1]DIMEVET - University of Bologna, Ozzano dell'Emilia, Bologna, Italy, [2]Department of Animal and Food Sciences, University of Delaware, Newark, DE, USA, [3]Vetagro S.p.A., Reggio Emilia, Italy, [4]Vetagro Inc., Chicago, IL, USA.*

To better study host-microbial interactions or stressful conditions that affect the porcine intestinal epithelium *in vitro* it is possible to use porcine apical-out enteroids, that better represent the intestinal epithelium compared with enterocytes monolayers or basal-out organoids. Botanicals are an interesting class of bioactive compounds with antioxidant and anti-inflammatory properties that help to support intestinal health. The aim of this study was to investigate the ability of a thymol-based blend of botanicals (BOT) to reduce the disruptive effects of lipopolysaccharide (LPS)-induced inflammation on apical-out porcine enteroids. Intestinal crypts were obtained from

35-day-old pigs' jejunum and they were used to generate apical-out enteroids. Those were then challenged with LPS for 6 h, in the absence or presence of BOT. The FD4 paracellular permeability and gene expression were then evaluated. Apical-out porcine enteroids responded to the LPS challenge by showing a 70% increase in FD4 paracellular permeability, a 2-fold increase in IL1β, IL8, and mucin 4 (MUC4) levels ($P < 0.01$), and a 3-fold increase in IL6 mRNA expression ($P < 0.01$). Moreover, a decrease in zonula occludens 1 (ZO1) and occludin (OCCL) levels was observed. BOT showed beneficial properties in reducing all the effects connected to the LPS challenge. The FD4 permeability was decreased by 35% ($P < 0.05$), and IL1β, IL6, IL8, and MUC4 expression were significantly reduced compared with the challenged control. Moreover, BOT increased the ZO1 and OCCL levels by 50% ($P < 0.05$), acting as a barrier-reinforcing agent. The BOT showed interesting anti-inflammatory properties, being able to maintain epithelial integrity thus reducing LPS-induced damage *in vitro*. This thymol-based blend has the potential to be further investigated as a feed additive to improve the pigs' ability to overcome stressful phases during their lifecycle.

Key Words: apical-out porcine enteroids, botanicals, inflammation, anti-inflammatory

Session 4

114 Tannins in poultry production. M. E. Fernandez Miyakawa*[1], [1]*INTA, Hurlingham, Buenos Aires, Argentina,* [2]*CONICET, Buenos Aires, Argentina.*

Tannins are a complex group of polyphenolic compounds found in a wide range of plant species that can precipitate proteins. These molecules are secondary metabolites of plants, which are found seeds, bark, wood leaves, and fruit skins and play important role in many natural ecological processes, such as herbivore defense, litter decomposition, nutrient cycling, nitrogen sequestration, and microbial modulation. The physical and chemical properties of tannins are particular of each plant species, the harvest region, and the extraction process. From chemical classification, plant tannins are grouped in hydrolyzable tannins and condensed tannins. Harmful nutritional consequences have been attributed to tannins because they can precipitate proteins, inhibit digestive enzymes, and decrease the utilization of vitamins and minerals. However, it highly depends on tannin concentration. The use of tannins in poultry production sector show favorable results, but the mechanism by which tannins promote growth and health in the monogastric animals are not clear. In general, tannins show variable antimicrobial, antiparasitic, antiviral, antioxidant, anti-inflammatory activities, and intestinal and environmental microbiota modulation. The data produced in the last few years about tannins and animal production provide an arena for discussion of each one of these actions potentially providing positive, negative, or neutral effects on productive efficiency, nutrition, gut health, and disease challenge. The ultimate improvement in performance arises because the sum of differences between these complex effects of tannins on nutrient digestion, microbial communities, and host response.

Key Words: tannins, poultry, health, production

115 Epigenetic regulation of host defense peptide synthesis. M. A. Whitmore and G. Zhang*, *Oklahoma State University, Stillwater, OK, USA.*

Host defense peptides (HDP) are an integral part of the first line of defense against infections. Modulation of HDP synthesis has emerged as a promising host-directed approach to fight against infections. We have identified several classes of epigenetic compounds capable of inducing HDP gene expression. The objective of this study is to explore a possible synergy in HDP induction among different classes of epigenetic compounds as an antibiotic-free approach to infectious disease control and prevention. Chicken macrophage cell lines and peripheral blood mononuclear cells were treated with or without structurally distinct histone deacetylase inhibitors (HDACi), histone methyltransferase inhibitors (HMTi), or DNA methyltransferase inhibitors (DNMTi) individually or in combinations, followed by analysis of the expression levels of HDP and cytokine genes as well as the major genes invovled in barrier function. We found that a combination of HDACi and HMTi or HDACi and DNMTi showed a strong synergy to induce the expression of multiple HDP genes. Tight-junction proteins such as claudin 1 were also synergistically induced by HDACi and HMTi, whereas LPS-induced *IL-1*β gene expression was suppressed. Overall, we conclude that HDP genes are regulated by epigenetic modifications. Strategies to increase histone acetylation while reducing DNA or histone methylation exert a synergistic effect on HDP induction and, therefore, have potential for the control and prevention of infectious diseases without relying on antibiotics.

Key Words: host defense, innate immunity, epigenetics, antibiotic alternatives, poultry

116 Software for microbiome and untargeted metabolomics in biomarker discovery. B. Shannon*, *BioRankings, St. Louis, MO, USA.*

Microbiome, untargeted metabolomics, and other -omics data has high potential for changing how biomarker discovery research is done in gut health in production of food animals. Although much research has been done and published in this area, the difficulty of analyzing this data and high risk of false positives has slowed down product development. BioRankings believes biostatistics, and not just bioinformatics, is needed to improve research and development, identify actionable biomarkers, and generate reproducible results. Building on decades of biostatistical analysis of -omics data in basic life sciences research and development, BioRankings provides statistical software that is unbiased, fast, and supports a wide range of experimental designs (e.g., one-way and multiway ANOVA, repeated measures, batch effects) and analyses (e.g., hypothesis testing, mean and confidence interval estimation). In this talk we show examples of software-as-a-service tools for untargeted metabolomics peak detection, microbiome hypothesis testing, and multi-omics regression analysis. Users can try this software for free with their data.

Session 5

117 Comparative efficacy of different natural products based on liquid Quillaja extract administered in the feed for the control of necrotic enteritis in chickens. S. Decap*[1], V. Tapia[1], R. Navarro[1], and B. Lumpkins[2], *[1]Plantae Labs SpA, Santiago, Chile, [2]Southern Poultry Research, Athens, GA, USA.*

Necrotic enteritis (NE) is one of the most concerning diseases in the poultry industry. Economic losses of NE ascend up to 40% of commercial broilers and are associated with the effect of *Clostridium perfringens* (CP) overgrowth in productivity. Treatment for NE administered in drinking water is more advantageous than feed treatment. Therefore, it is important to offer new natural solutions. The study was conducted at Southern Poultry Research Inc. using 0- to 28-day-old Cobb 500 male chicks. There were 72 cages with 8 birds/cage and 9 cages/treatment. On d 14, all birds except control were orally inoculated with ~5,000 oocysts of *Eimeria maxima* per 1-mL dose and from 19 to 21 d received an inoculum of ~108 cfu/mL CP. Lesion score (0 to 3 score), feed intake, body weight gain (BWG), and feed conversion ratio (FCR) were recorded. Blood samples for FITC-d analysis were taken on d 21 (2 birds/cage). Groups were control (CON), control/challenged (POS), Maxiban (MAX), Quillaja-oregano emulsion (QOE1: 0.12 g/L, ratio 6:1), Quillaja-oregano emulsion (QOE2; 0.3 g/L, ratio 1:6), and Quillaja liquid solution (QLS; 0.06 g/L). The NE challenge had a clear effect on performance as observed by the lower BWG and FCR in POS versus CON; between 14 and 20 d, birds in POS had a 65% reduction in BWG versus CON. Compared with untreated birds (POS), QLS had a significant reduction (16%) and increase (23%) in FCR and BWG, respectively. All treatments had lower FCR than POS, however, QLS and MAX had the lowest values. Following the challenge, between 14 and 28 d, there was a noticeable difference in BWG of both QLS and QOE2 groups versus POS with an increase of 37.8 and 13.6%, respectively. The average lesion score in MAX and QLS was 51.1 and 44.6% lower than POS, respectively. The FITC-d was 10.4% lower than POS in QOE2, whereas QOE1 group showed a 9.4% reduction in FITC-d versus POS. In conclusion, there was a clear effect of *Eimeria* and *C. perfringes* challenge on the performance of birds. The most promising results were obtained using QLS and QOE2 as natural treatments.

Key Words: Quillaja extract, essential oil, necrotic enteritis, leaky gut, chickens

118 Describing the cecal microbiota development in broilers when fed varying levels of a yeast culture product. L. A. Wythe*[1], D. K. Dittoe[1], A. Scheaffer[2], and S. C. Ricke[1], *[1]Meat Science and Animal Biologics Discovery Program, Department of Animal and Dairy Science, University of Wisconsin, Madison, WI, USA, [2]Harvest Fuels, Walhalla, ND, USA.*

The objective was to evaluate a yeast culture product, ProBiotein® (PB), on the cecal microbiome development of Ross 308 males fed PB supplemented diets for 42 d. Diets were offered *ad libitum* and consisted of an industry-standard basal diet (CON), basal + 0.0% PB (0.0PB)., basal + 0.5% PB (0.5PB), or basal + 0.75% PB (0.75PB). Chicks were randomly assigned to batteries using an RCBD on d 0 (n = 400, n = 10, k = 4, r = 2) and moved to floor pens on d 14 following the same blocking patterns. Starter, grower, and finisher phases were fed in 14-d intervals. On d 14, 28, and 42, 10 birds per treatment were euthanized, and cecal digesta was collected. Genomic DNA were extracted, and the V4 region of the 16S rRNA gene was sequenced on an Illumina MiSeq. Sequencing data were analyzed in QIIME2–2022.2. Treatment, block, day effects, and the interactions between were analyzed on α and β diversity using ANOVA and ADONIS with significance determined at $P < 0.05$. Pairwise differences were determined using Kruskal-Wallis (α) or ANOSIM (β) ($P < 0.05$, Q <0.05). Microbiome maturity was evaluated per treatment using MAZ scores ($P < 0.05$). Significantly different taxonomical relative abundances were determined using ANCOM. Significant interactions across α and β diversity revealed shifts in cecal microbiota due to both treatment and day ($P < 0.05$, Q <0.05). The treatment × day ANCOM revealed 51 significantly different relatively abundant taxa at the genus level (120 < W <160, $P < 0.05$). The MAZ scores showed a steady microbial maturity from d 14 to 42 for birds fed CON. Maturity for those fed 0.2PB and 0.5PB decreased sharply at d 28 but increased sharply toward similar scores as those fed CON. The MAZ scores for those fed 0.75PB decreased steadily from d 14 through d 42. Results show that microbiota development over time can be modulated by dietary treatments and should be accounted for when evaluating feed amendments. In conclusion, PB was shown to influence microbiota development and may be a useful feed additive in promoting microbial diversity in the ceca.

Key Words: microbiome, microbiota, yeast fermentate, broilers

119 Types and dietary levels of resistant starches modulated growth performance and intestinal environment in broiler chickens. I. W. Oluseyifunmi* and O. A. Olukosi, *University of Georgia-Athens, Athens, GA, USA.*

In a 21-d study, 480 Cobb 500 (off-sex) male chicks were used to evaluate the effects of feeding different types and levels of resistant starches (RS) on growth, digesta pH,

digestive tract morphometry and jejunal histomorphology in broiler chickens. The birds were allocated to 10 treatments in a 3 × 3+1 factorial arrangement. The factors were 3 RS types (RST): banana starch (BS), raw potato starch (RPS), and high-amylose corn starch (HCS); each at 3 levels (RSL) 25, 50 or 100 g/kg plus a corn-SBM control. Birds and feed were weighed on d 0, 8 and 21. On d 21, samples of tissues and digesta were used for morphometry, histomorphology, and digesta pH determination. There were no significant main effects or RST × RSL for d 0 to 21 weight gain but there was a tendency for RST × RSL ($P = 0.087$) for feed intake. Feed intake was higher ($P < 0.01$) for birds receiving RPS compared with the control whereas FCR tended to be higher ($P = 0.056$) for birds receiving BS compared with the other RST or control. There were no RST × RSL or RST main effect for digesta pH in the small intestine or ceca but jejunal pH tended to be lower ($P = 0.069$) at 100 g/kg RSL. RST × RSL was significant ($P < 0.05$) for relative ileal length with general increase in ileal length with increasing level of all RST except HCS. There was trend for RST × RSL ($P = 0.096$) for relative ceca weight due to increase of ceca weight with increasing BS and RPS levels but not with HCS. Relative duodenum weight at RSL 50 and 100 g/kg tended to be greater ($P = 0.085$) than 25 g/kg level or the control. Jejunal villi were longer ($P < 0.05$) for all RST compared with control but longest villi were in birds receiving HCS diet; and villi height tended to increase ($P = 0.058$) with increasing RSL. Birds receiving RPS and HCS had deeper ($P < 0.01$) crypt than birds on control and BS. The current study showed that the effects of RS on growth performance (mainly FCR differences modulated by feed intake effects), gut morphology and villi and crypt characteristics depend on both the type and level of RS in the corn-soybean meal diet.

Key Words: resistant starches, growth, pH, histomorphology, chickens

120 Rumen microbial cluster identification and its influence on rumen metabolites and growth performance of young goats. D. Wang[1], G. Tang[1], L. Wang[1,2], X. Chen[2], J. Yao[1], and Y. Cao*[1,2], *[1]Northwest A&F University, Yangling, Shaanxi, China, [2]Harvard Medical School, Boston, MA, USA.*

The microbial clusters in the intestine (enterotypes) have been found to relate to host metabolism and health. It is the first time that ruminal bacterial clustering in young goats is studied in the present experiment. The ruminal microbiome in 99 6 mo-old goats was analyzed and identified to 2 enterotypes: cluster 1 (n = 38) was dominated by genus *Prevotella* (P-cluster), and cluster 2 (n = 61) was dominated by *Ruminococcus* (R-cluster). Compared with P-cluster goats, R-cluster goats had greater growth rates, the concentrations of propionate, butyrate, and 18 free amino acids, and the proportion of unsaturated fatty acids, but lower acetate molar percentage, the acetate to propionate ratio and several odd and branched chain and saturated fatty acids in rumen fluid. The ruminal microbiome composition differed significantly between 2 enterotypes. At the genus level, several members of *Firmicutes*, including *Ruminococcus*, *Oscillospiraceae* NK4A214 group and *Christensenellaceae* R-7 group were significantly higher in R-cluster, whereas *Prevotellaceae* members, such as *Prevotella* and *Prevotellaceae* UCG-003, were significantly higher in P-cluster. Spearman correlation analysis showed that those R-cluster-enriched bacteria have negative correlations significantly with those P-cluster-enriched bacteria. Moreover, we found the concentrations of propionate, butyrate and free amino acids, and the proportion of unsaturated fatty acids were positively correlated with those R-cluster-enriched bacteria. The concentrations of acetate, acetate to propionate ratio and the proportion of saturated fatty acids and odd and branched chain fatty acids were positively correlated with those P-cluster-enriched bacteria.

Key Words: rumen, microbial cluster, metabolites, growth performance, goats

Poster Session

P100 Microbial interventions to improve gut health in neonatal calves. R. Nakandalage*[1,2], L. L. Guan[1], and N. Malmuthuge[2], [1]*University of Alberta, Edmonton, AB, Canada,* [2]*Agriculture Agri-Food Canada, Lethbridge, AB, Canada.*

Scour is one of the gastrointestinal diseases in newborn calves and one of the main reasons for antimicrobial usage in the dairy industry. Previous studies reported that colostrum feeding not only transfers maternal antibodies to neonatal calves but also enhances the colonization of beneficial microbes in the gut. Failure to transfer passive immunity (FTPI) via colostrum, increases the risk of developing enteric infections such as scours during early life, a common concern in the dairy industry. Therefore, it is essential to identify effective approaches to improve calf gut health that increase the resilience to enteric infections and reduce antimicrobial use in the industry, especially during FTPI. Microbial intervention during early life has been suggested as a potential effective management approach to create a long-term effect on animal health. However, knowledge is limited on early life microbial interventions on gut microbiota colonization and gut physiology in calves with FTPI. This study hypothesizes that bovine-derived probiotics or prebiotics supplementation during early life can create a favorable environment for the colonization of beneficial microbiota and improve host resilience to enteric infections when calves experience FTPI. In this 35-d study, newborn calves (n = 50) will be assigned to one of the 2 treatment groups: (1) suboptimal transfer immunity (SO; n = 40) with 1/4 of their recommended IgG and (2) optimal group (n = 10) with the recommended level of IgG via colostrum within 2 h of birth. Calves in SO will be randomly assigned to one of the 4 treatment groups during the first 2 weeks of life: (1) probiotic (10^9 cfu; *Bifidobacterium*), (2) prebiotic (80 g; resistant potato starch), (3) synbiotic, and (4) control. We will study the dynamics of gut microbiota, oxidative stress, and metabolites to explore the gut health of calves during the first 5 weeks of life. This study will help to develop effective management strategies to improve gut health while reducing preweaning calf morbidity and mortality.

Key Words: dairy calves, gut health, microbial interventions

P101 Digital PCR as a new highly sensitive method in chicken cytokine profiling. G. Giovagnoni[2], F. Perry*[1], B. Anderson-Coughlin[1], K. Kniel[1], B. Tugnoli[3], A. Piva[1,3], E. Grilli[2,4], and R. Arsenault[1], [1]*Department of Animal and Food Sciences, University of Delaware, Newark, Newark, DE, USA,* [2]*DIMEVET, Dipartimento di Scienze Mediche Veterinarie, Università di Bologna, Bologna, Italy,* [3]*Vetagro S.p.A., Reggio Emilia, Italy,* [4]*Vetagro Inc., Chicago, IL, USA.*

Chicken cytokine profiling is an established tool used to study the status of the immune system of animals for research and diagnostic purposes. Real-time quantitative PCR (qPCR) is now the simplest and fastest method for gene expression analysis. However, the major disadvantage of this technique is its limited sensitivity in the detection of low expressed genes. Digital PCR (dPCR), on the other hand, is a more recent technique, based on a highly sensitive end point absolute quantification of gene copy number. The aim of this study was to compare cytokine expression in chicken HD-11 cells infected with *Salmonella Enteritidis* using both qPCR and dPCR. For 6 h, cells were infected with the bacterium alone (Positive Control) or infected and treated with different antibiotics or botanicals, a group without infection and treatments was included (Negative Control; n = 3 all groups). For the qPCR, cDNA was reverse-transcribed and the analysis was performed using PowerUp SYBR Green Master Mix kit. For the dPCR, the QIAcuity OneStep Advanced Probe Kit was used to perform the analysis on RNA samples. The IL1β, IL6, IL10, and IFNγ cytokines was investigated. To compare the results of the 2 methods, data were expressed as 2^(Treatment Ct – Negative Control Ct) for qPCR or as the difference of copies/uL between Treatment and Negative Control for dPCR. Data were analyzed with one-way ANOVA followed by Tukey's multiple comparisons test. Differences were considered significant at $P \leq 0.05$. Statistical significances for IL10 and IFNγ were equivalent comparing the 2 methods. For IL1β, no differences were registered with qPCR, whereas dPCR data indicated statistical significance among groups ($P = 0.003$). The dPCR provided several significant differences that the qPCR was not able to highlight for IL6. This could be explained by the greater sensitivity of dPCR in detecting copy numbers of *IL6* gene in the Negative Control group, because the qPCR was not able to determine the Ct. Also the IFNγ Ct of several samples resulted as undetermined in qPCR analysis, in contrast to dPCR. dPCR can therefore be a candidate technique to replace qPCR in chicken cytokine profiling thanks to its high sensitivity.

Key Words: dPCR, cytokine profiling

P102 Citrus based additive characterization to explain microbiota modulation of peripartum sows and beneficial consequence on performances. S. Cisse*[2,3], R. Cornet[1,3], E. Belz[2], M. E. A. Benarbia[2,3], O. Zemb[4], and D. Guilet[1,3], [1]*EA 921 SONAS, Beaucouzé, Maine et Loire, France,* [2]*Nor-Feed SAS, Beaucouzé, Maine et Loire, France,* [3]*Joint Lab ANR FeedInTech,* [4]*UMR GenPhySE, INRAE, Toulouse, Haute-Garonne, France.*

Farrowing is a critical period for sows. This stage is very suitable for digestive disorder such as constipation. Most of the time, these troubles are closely linked to dysbiosis and can have harmful consequences on animals. To manage these situations, intestinal microbiota modulation could be an effective solution. The aim of this study was to evaluate the effect of a standardized citrus extract (SCE, Nor-Spice® AB, Nor-Feed SAS) on peripartum sows and their offspring. The SCE has also been characterized to better understand the observed effect on sows. Fifty sows on peripartum were divided into 2 groups: (1) a control (CTL) group (23 sows) fed with a standard diet; and (2) an SCE group (27 sows) fed with standard diet and supplemented with 2,500 ppm of SCE, 10 d before farrowing and 5 d after. Sows' performances (feed intake, litter weight gain and transit resumption) and microbiota were monitored. In parallel, SCE compounds were identified using GC-MS and LC-MS (dereplication). Results showed that SCE supplementation allowed to increase sows' feed intake compared with CTL group. The interval between farrowing and first dejection was also reduce in the SCE group, which indicate a better and faster transit resumption after farrowing. Moreover, piglets litter weight gain between 24 h and 7 d after farrowing was higher in SCE group compared with CTL group. The SCE characterization allowed to identify pectic oligosaccharides as SCE major components and confirm the presence of 30 secondary metabolites including citric acid, caffeic acid, eriocitrin, hesperidin and naringin. These compounds are well known for their positive effect on different compartments of the gut and microbiota, according to the literature. In conclusion, SCE supplementation showed beneficial effects on *peripartum* sows' performances and welfare. These effects were correlated with sow's microbiota modulation. The SCE characterization allowed the identification of molecules that explain in part the observed effect on sows. Further studies will be necessary to confirm the role of these molecules.

Key Words: citrus extract, sows, farrowing, characterization, microbiota management

P103 Differential growth of *Campylobacter jejuni* in mixed populations of porcine fecal, bovine fecal, or bovine ruminal microbes. R. C. Anderson*, M. E. Hume, N. A. Krueger, R. B. Harvey, T. L. Poole, and T. L. Crippen, *USDA, Agricultural Research Service, Southern Plains Agricultural Research Center, Food and Feed Safety Research Unit, College Station, TX, USA.*

Campylobacter is a leading bacterial cause of human foodborne illness in the United States. *Campylobacter* are found more frequently and at higher numbers in the intestinal tract than in the rumen. This suggests *Campylobacter*, which occupy an amino acid-fermenting niche, may be less competitive in the rumen due to resident populations of competent amino acid-fermenting and ammonia-assimilating microbes. To test this hypothesis, rumen and fecal samples, collected freshly from a cannulated cow, and freshly collected feces from a market hog were each diluted 10^{-4} in anaerobic buffer to deplete endogenous substrate. The populations were tested individually or jointly by inoculating each of the diluted suspensions alone (0.2 mL) or in pairs (0.1 mL each) to tubes containing 10^5 cfu of *C. jejuni* in Bolton broth augmented with 0.2% glucose, cellobiose, and xylose. Total anaerobes measured before inoculation were 4, 3, and 4 $\log_{10}$ cfu/mL in rumen, bovine fecal, and porcine fecal mixtures, respectively and contained no wildtype *Campylobacter*. Results revealed that after 48 h of anaerobic incubation (39°C), *C. jejuni* were lowest ($P < 0.05$) in cultures inoculated with suspensions of rumen microbes alone or rumen:bovine fecal mixtures (5.7 and 6.4 $\log_{10}$ cfu/mL, respectively). *Campylobacter jejuni* were highest ($P < 0.05$) in cultures inoculated with swine fecal suspensions alone or control *C. jejuni* cultured without any of the suspensions (8.0 and 8.3 $\log_{10}$ cfu/mL, respectively) and intermediate in the other tested cultures (7.2 to 7.6 $\log_{10}$ cfu/mL). Ammonia accumulations were higher ($P < 0.05$) in *C. jejuni* cultures inoculated with porcine fecal:bovine fecal, rumen:porcine fecal mixtures and porcine fecal suspensions alone (8.2, 8.1 and 6.3 µmol/mL, respectively) than in the other cultures (<1.9 µmol/mL). At 48 h, total anaerobes in cultures inoculated with the mixed gut microbes differed modestly ($P < 0.05$), ranging from 7.1 to 7.7 $\log_{10}$ cfu/mL. Results warrant further study to characterize population differences in amino acid metabolism that may affect growth of *C. jejuni* in rumen and fecal sources.

Key Words: *Campylobacter*, food safety, rumen, swine

P104 Multistrain probiotics positively affect the growth performance, meat quality and alter the fecal microbiota in broiler chickens. C. B. Lim*, Q. Q. Zhang, and I. H. Kim, *Department of Animal Resource and Science, Dankook University, Cheonan, South Korea.*

The study aimed to explore the effectiveness of incorporating *Bacillus subtilis, Bacillus coagulans, Bacillus licheniformis and Clostridium butyricum* on the growth performance, nutrient digestibility, fecal toxic gas emission, fecal microbiota, and meat quality of broiler chickens. A total of 720 one-day-old Ross-308 broilers with an initial average BW of 42.73 ± 2.05 g were randomly allotted to 4 treatments (10 replicate pens/treatment and 18 broilers/pen). The experiment lasted 35 d. The broilers were fed a basal diet or a basal diet containing 0.05%, 0.1%, and 0.2% multistrain probiotics. Experimental data were analyzed using GLM procedure of SAS (SAS Institute Inc.) with Duncan's multiple range

test. Probability level $P < 0.05$ was considered statistically significant and $P < 0.10$ as a trend. A linear increase in body weight gain was observed ($P < 0.05$) with the increasing levels of probiotics from d 1 to 7, d 8 to 21, and d 1 to 35 ($P < 0.05$). Additionally, there was a dose effect of probiotics on increased feed intake ($P = 0.030$) during d 1 to 35. There was no difference observed in the nutrient digestibility of dry matter, nitrogen, and gross energy. However, dietary probiotics supplement increased ($P < 0.05$) the *Lactobacillus* and tended to reduce *Escherichia coli* counts in the broiler. The pH value and water holding capacity ($P = 0.0694$) tended to increase ($0.05 < P < 0.1$) as the probiotics dose increased. Taken together, multistrain probiotic plays vital roles in improving BWG and altering microbiota, and improving meat quality.

Key Words: multistrain probiotic, growth performance, meat quality, fecal microflora, broiler

P105 Effect of Jefo P(OA+EO+PC) to control coccidiosis and reduce intestinal lesions. C. Roy*, L. Lahaye, H. Salgado, R. Scott-Delaunay, and E. Santin, *Jefo Nutrition Inc., St-Hyacinthe, QC, Canada.*

Coccidiosis is a common enteric disease caused by *Eimeria* and is a big challenge in the poultry industry due to its negative effect on broilers' performance and economics worldwide. This parasite causes inflammation in the gastrointestinal tract and affects microbial community by dysbiosis, which causes losses and lacks in performance and health. Controlling the dysbiosis of microbial community is a part of the strategy to reduce the negative effects of coccidiosis in broilers. A total of 1,080 male broiler chickens (Ross 308) were raised until 35 d of age and distributed into 3 treatments with 12 replicates (floor pens with 30 birds/pen). Treatments were (1) nonchallenged control (NC), (2) challenged control (CC), and (3) CC + Jefo P(OA+EO+PC) (700 g/t). Challenge birds were inoculated with 200k oocysts (*Eimeria acervulina* 73%; *Eimeria maxima* 7% and *Eimeriatenella* 30%) on d 18. At d 23, a jejunum sample from one bird per pen was collected and analyzed by I See Inside (ISI®) scoring system methodology. Parametric data were submitted to ANOVA and Tukey's test for the means with a significant difference ($P < 0.05$). Nonparametric data were submitted to the Kruskal-Wallis test ($P < 0.05$). Results showed that at d 23, inflammatory cell infiltration on jejunum epithelium and presence of oocysts were significantly lower with the use of Jefo P(OA+EO+PC) compared with birds who had coccidiosis challenge ($P < 0.001$). These results suggest that the supplementation of Jefo P(OA+EO+PC) promotes a better intestinal health and faster recovery from a coccidiosis challenge.

Key Words: coccidiosis, broilers, additives, gut health

P106 Use of a fermented product with saponins to reduce intestinal lesions during coccidiosis challenge in broilers. H. H. Salgado*, L. Ludovic, C. Roy, R. Scott-Delaunay, and E. Santin, *Jefo Nutrition Inc. Saint-Hyacinthe, Quebec, Canada.*

The negative effects in performance due to the intestinal lesions caused by *Eimeria* are well documented in poultry. Ionophoric coccidiostats such as Salinomycin (SAL) are widely used as a supplement in poultry diets to control coccidiosis. However, anticoccidial resistance has prompted saponins (SAP), a plant-derived compound as an alternative to prevent and control this disease. Therefore, the objective of this study was to compare the effectiveness of 2 SAP extract-based products as alternatives to SAL to reduce intestinal lesions in coccidiosis challenge. A total of 624 male broiler chickens (Ross 308) were randomly allocated to the 3 treatments which consisted in the supplementation of the 3 commercial products to a basal diet (BD): (1) BD + 0.5 g of SAL/kg of diet (control), (2) BD + 0.5 g of Jefo-SAP/kg of diet, (3) BD + 0.5 g of SAP-B/kg diet. On d 18 all birds were gavaged with live *Eimeria* (*E. acervullina*, *E. maxima*, and *E. tenella*). At d 23, one bird per pen was slaughtered and lesions in the gastrointestinal tract were evaluated by I See Inside scoring system methodology (ISI®). Data were submitted to ANOVA. Body weight at d 35 was similar among control birds and birds supplemented with Jefo-SAP (,2191.9 g and 2,117.4 g, respectively $P > 0.05$), but not for birds supplemented with SAP-B (2,106.9 g, $P < 0.05$). The FC at d 35 was similar among control birds and Jefo-SAP birds (1.52 and 1.54, respectively, $P > 0.05$), but SAP-B was higher than control (1.57; $P < 0.05$). The ISI score of lesions in the gastrointestinal tract was not different among SAP and control; however, the lesion score in the intestine was numerically lowest in birds supplemented with Jefo-SAP (3.5 vs. 5.8 and 4.5 for control and SAP-B, respectively). In conclusion, the fermented product with SAP can be used as an alternative to coccidiostats to prevent GAT lesions in coccidiosis infection while improving BW and FC in broilers.

Key Words: *Eimeria*, broilers, additives

P107 Effects of supplemental encapsulated butyric acid and zinc on rumen morphometrics and small intestine histology of finishing lambs abruptly transitioned to a grain-based diet. F. L. Francis*[1], B. B. Grimes[1], T. L. Maia Ribeiro[1], E. R. Gubbels[1], D. Lafleur[2], J. E. Hergenreder[2], and Z. K. Smith[1], *[1]South Dakota State University, Brookings, SD, USA, [2]Kemin Industries, Inc., Des Moines, IA, USA.*

The objective of this study was to determine if encapsulated butyric acid and zinc (BZ) fed to feedlot lambs abruptly

transitioned to a grain-based diet influences rumen morphometrics and small intestinal histology. Polypay wethers (n = 84) were assigned to dietary treatment (CON: 0 g/kg BZ diet DM; BZ: 2 g/kg diet DM) in a randomized complete block design. Wethers were abruptly transitioned from a diet based upon grass hay (0.95 Mcal/kg NEg) to a finishing diet based upon whole corn (1.33 Mcal/kg NEg) upon study initiation. Four wethers were harvested on day (d) 0 to serve as a baseline, and the remaining 80 lambs were serially harvested on d 7, 14, 21, 56, and 63 (8 CON and 8 BZ per harvest date) for the collection of rumen, duodenum, and ileum samples. Rumen samples were stored in a 70% ethanol solution for storage until evaluation for papillae count, mean papillae area, and calculated rumen wall absorptive surface area. Duodenal and ileal samples were fixed in 10% neutral buffered formalin, embedded in paraffin, and stained with hematoxylin and eosin for microscopic evaluation of villus height, crypt depth, villi-to-crypt ratio, and mucosal thickness. Data were analyzed with individual wether as the experimental unit and fixed effects of treatment, harvest date, and their interaction. No treatment × day interactions ($P \geq 0.23$) were observed for any variables. Additionally, BZ did not appreciably affect ($P \geq 0.26$) any measures. A positive linear effect ($P \leq 0.04$) of harvest date was observed for rumen papillae area, rumen papillae count, calculated rumen wall absorptive surface area, ileal villi height, ileal villi-to-crypt-ratio, and duodenal mucosal thickness. Supplemental dietary BZ had no effect on rumen morphometrics or small intestine histology in lambs abruptly transitioned from a forage- to grain-based diet. However, increasing days on a grain-based diet appears to affect these measures.

Key Words: butyric acid, histology, intestine, rumen, zinc

P108 Broiler gastrointestinal tract responses to commercial diets containing different levels of a yeast fermentate. L. A. Wythe*[1], D. K. Dittoe[1], A. Scheaffer[2], and S. C. Ricke[1], *[1]Meat Science and Animal Biologics Discovery Program, Department of Animal and Dairy Science, University of Wisconsin, Madison, WI, USA, [2]Harvest Fuels, Walhalla, ND, USA.*

The objective was to evaluate a yeast culture product, ProBiotein® (PB), on the gastrointestinal (GIT) morphology of Ross 308 males fed PB supplemented diets. Treatments consisted of a basal diet (Con), basal+0.2% PB (0.2PB), basal+0.5% PB (0.5PB), or basal+0.75% PB (0.75PB). Birds were randomly assigned to batteries on d 0 (n = 400, n = 10, k = 4, r = 2) and moved to floor pens following the same blocking design on d14. Birds were offered feed *ad libitum* with a starter, grower, and finisher phase, each 14 d. On d 14, 28, and 42, body weight, GIT weight, length, and pH were measured. Weight was reported as relative to body weight. Cross sections of the duodenum, jejunum, and ileum from d 14 were evaluated for villi height, crypt depth, and their ratio. Morphology data were analyzed using a linear mixed effect model (mixed effect = block) with Dunnett's comparison with significance at $P \leq 0.05$ (control = Con). Histology data were analyzed using a linear model ($P \leq 0.05$). There were no differences found histologically. There were treatment × day interactions on gizzard, jejunal, ileal, and small intestine weights; and for duodenal, ileal, and small intestine lengths ($P < 0.05$). There was a treatment effect on gizzard pH and a treatment × day interaction on the cecal pH ($P < 0.05$). On d14, jejunal, ileal, and small intestine weights were greater in birds fed diets with 0.75PB compared with those fed Con ($P < 0.05$). Small intestine length was longer in birds fed 0.75PB compared with those fed Con ($P < 0.05$). On d28, gizzard pH was lower in birds fed diets with 0.75PB than in those fed Con ($P < 0.05$). On d42, duodenal lengths were lower in birds fed diets supplemented with 0.75PB compared with those fed Con ($P < 0.05$). On d42, birds fed diets with 0.75PB had longer ileums than those fed Con ($P < 0.05$). Cecal pH was higher in birds fed all treated diets on d14 but was then lower on d28 and 42 compared with those fed Con ($P < 0.05$). Overall, birds fed diets supplemented with PB had lower GIT weights, longer GIT segments, and lower GIT pH. Supplementing ProBiotein® into a commercial poultry diet altered early intestinal development in chicks.

Key Words: yeast fermentate, poultry, gastrointestinal physiology, pH

P109 Effects of different stocking density and phytogenic feed additives dosage levels on growth performance, nutrient digestibility, blood profile, and behavior change of growing-finishing pigs. H. Cho*[1], M. Song[3], J. Lee[2], H. Oh[1], Y. Kim[1], S. Jang[1], J. An[1], Y. Go[1], D. Song[1], S. Cho[4], D. Kim[4], M. Kim[4], H. Kim[5], and J. Cho[1], *[1]Chungbuk National University, Cheongju-si, Chungcheongbuk-do, Republic of Korea, [2]University of Georgia, Athens, GA, USA, [3]Chungnam National University, Daejeon, Republic of Korea, [4]Eugenebio, Suwon-si, Gyeonggi-do, Republic of Korea, [5]Dankook University, Cheonan-si, Chungcheongnam-do, Republic of Korea.*

This study was to investigate the effects of different phytogenic feed additives (PFA) dosage levels in grower finishing pigs with stressed by high stocking density. A total of 72 mix sexed 12-week growing pigs [(Landrace × Yorkshire) × Duroc] with initial body weight of 49.28 ± 4.58 kg were used for 8 weeks. There were 3 replicate pens in each treatment, with 3 pigs per pen. The dietary treatment consisted of basal diets in animal welfare density (NC, negative control), basal diet in high stocking density (PC, positive control), PC + essential oil (T1), PC + 0.08% essential oil (T2), PC + 0.10% bitter citrus extract and essential oil (T3), PC + 0.2% bitter citrus extract and

essential oil (T4), PC + 0.05% grape pomace extract (T5), PC + 0.10% grape pomace extract (T6). The reduction of space allowance decreased ($P < 0.05$) growth performance such as average daily gain, feed efficiency and nutrient digestibility such as dry matter, crude protein, and gross energy. Also, the fecal score of PC groups increased ($P < 0.05$) compared with other treatments. Basic behaviors (feed intake, standing, lying) were inactive ($P < 0.05$) and singularity behavior (biting) was increased ($P < 0.10$) under high stocking density. There was no difference in blood profile. However, the supplementation of PFA alleviated the negative effects such as reducing growth performance, nutrient digestibility, some increasing stress indicators in blood and animal behavior. In conclusion, the supplementation of PFA improved the health of pigs with stress by high stocking density and low dosage of bitter citrus and essential oil (T3) is the most effective.

Key Words: pig, plant extract, dosage, stocking density, stress

P110 Phytogenic feed additives alleviate pathogenic *Escherichia coli*-induced intestinal damage through improving barrier integrity and inhibiting inflammation in weaned pigs. S. Chang*[1], M. Song[2], J. Lee[3], H. Oh[1], Y. Kim[1], J. An[1], Y. Go[1], D. Song[1], H. Cho[1], S. Cho[4], D. Kim[4], M. Kim[4], H. Kim[5], and J. Cho[1], *[1]Chungbuk National University, Cheongju-si, Chungcheongbuk-do, Republic of Korea, [2]Chungnam National University, Daejeon, Republic of Korea, [3]University of Georgia, Athens, GA, USA, [4]Eugenebio, Suwon-si, Gyeonggi-do, Republic of Korea, [5]Dankook University, Cheonan-si, Chungcheongnam-do, Republic of Korea.*

This study was conducted to investigate the effect of each phytogenic feed additive (PFA) on the growth performance, nutrient digestibility, intestinal morphology, and immune response of weaned pigs challenged with *Escherichia coli*. A total of 63 4-week-old weaned pigs with initial body weight of 8.03 ± 0.43 kg were placed in individual metabolic cages, and then randomly assigned to 7 treatment groups. The 7 treatments were as follows: (1) NC: basal diet without *E. coli* challenge, (2) PC: basal diet with *E. coli* challenge,(3) T1: PC + 0.04% bitter citrus extract, (4) T2: PC + 0.01% microencapsulated blend of thymol and carvacrol, (5) T3: PC + 0.10% mixture of bitter citrus extract and microencapsulated blend of thymol and carvacrol, (6) T4: PC + 0.04% premixture of grape seed and grape marc extract, green tea and hops, (7) T5: PC + 0.10% fenugreek seed powder. The experiments progressed in 21 d, including 7 d before and 14 d after the first *E. coli* challenge (d 0). In the *E. coli* challenge treatments, all pigs were orally inoculated by dividing a total of 10 mL of *E. coli* F 18 for 3 consecutive days from d 0 postinoculation (PI). Compared with PC treatment, the PFA-added treatment groups significantly increased ($P < 0.05$) average daily gain and feed efficiency, and decreased ($P < 0.05$) the fecal score at d 0 to 14 PI. Tumor necrosis factor α was significantly lower ($P < 0.05$) in the PFA-added treatment groups except for T1 in d 14 PI compared with the PC treatment. T3 showed significantly higher ($P < 0.05$) villus height:crypt depth and claudin 1 expression in ileal mucosa, and significantly downregulated ($P < 0.05$) the expression of calprotectin. In conclusion, the addition of PFA in weaned pigs challenged with *E. coli* alleviated the negative effects of *E. coli* and improved growth performance. Among them, the mixed additive of bitter citrus extract and essential oils showed the most effective results, improving immune response, intestinal morphology, and expression of tight junctions.

Key Words: barrier integrity, immunity, phytogenic feed additive, postweaning diarrhea, weaned pig

P111 Survey of yeast populations in fermented feed across the United States. F. R. Mazza*, J. S. Thompson, and A. H. Smith, *Arm & Hammer Animal and Food Production, Waukesha, WI, USA.*

Yeasts are single-celled, facultative anaerobic eukaryotes that are prevalent in various fermented feeds. Yeasts can negatively affect silage quality by metabolizing available nutrients within the feed which leads to dry matter loss and decreased nutritive value, and by producing ethanol which can lead to off flavors in milk when consumed. Yeasts also oxidize lactic acid, which increases silage pH and promotes the growth of organisms such as mycotoxin-producing molds. Although wild yeasts can have a negative effect on feed quality, the prevalence and identities of these organisms in different crop types are not well studied. The objective of this survey was to examine yeast populations in fermented feed from various sites across the United States. From April 2019 to July 2022, 438 samples of 3 different crops (alfalfa haylage; n = 132), corn silage (CS; n = 272) and high moisture corn (HMC; n = 34) were collected from more than 130 farms across 16 states. Yeasts from these samples were enumerated on potato dextrose agar with 0.15% tartaric acid. Isolates were then harvested and DNA was extracted. Species were identified by sequencing the fungal ITS region. From the feed samples that were analyzed, yeasts were detected more often in HMC (76.5%) than alfalfa haylage (50.8%; $P < 0.05$). Yeasts were detected at higher levels in HMC compared with both alfalfa haylage and CS ($P < 0.0001$). The CS had the highest species diversity (14 species), with a Shannon-Weiner index value of 2.2, whereas HMC had the lowest diversity (4 species), with a Shannon-Weiner index value of 0.99. *Pichia kudriavzevii*, *Pichia fermentans*, and *Saccharomyces cerevisiae* were common species found across all 3 feed types. *P. fermentans* was the most prevalent species in both CS and alfalfa haylage, whereas *Wickerhamomyces anomalus* was the

most prevalent species found in HMC. Overall, the data gathered provides a better understanding of yeast levels and populations found in various fermented feeds across the United States. This knowledge can be used in future research to identify novel methods to inhibit yeast growth within fermented feed.

Key Words: yeast, silage, fermentation

P112 The effect of reduced whey protein and semi-moisture diet with different energy content on growth performance and nutrient digestibility in newly weaned piglets. C. B. Lim*, M. M. Hossain, and I. H. Kim, *Department of Animal Resource and Science, Dankook University, Cheonan, South Korea.*

This study aimed to investigate the effects of reduced whey protein and semi-moisture with varying calorie content diet on growth performance, nutrient digestibility in weaned piglets, and moisture content of different forms of feed. A total of 75 weaned piglets ((Landrace × Yorkshire) × Duroc, 28 d old, 7.17 ± 0.15 kg body weight) were randomly allocated into 3 treatment groups (5 replicate pens per treatment, each pen containing 5 weaned piglets; n = 25/treatment) for 35 d. The 3 treatments were: (1) Control (whey protein) by pellet, (2) Control (whey protein with energy modify) fed semi-moisture diet by extrusion, and (3) No whey protein with energy up fed semi-moisture diets by extrusion. Results revealed that the average daily gain, feed intake, and feed conversion ratio didn't change through the supplementation of semi moisture diet with different levels of whey protein and energy diet in weaned piglets. At wk 5 of the feeding trial, nitrogen digestibility was lower in energy-controlled and semi-moisture feed than in the control and no whey protein modified energy semi-moisture feed. Digestion of dry matter and energy was unaffected by diet energy or moisture. Semi moisture feed group contain less dry matter compared with the control group in both the second and fifth group. And moisture content in semi moisture feed with whey protein was lower than semi moisture feed without whey protein in second week which alter in fifth week. Here we found that semi moisture diet with different levels of whey protein and energy diet showed similar growth performance to control feed in weaned piglets. But extrusion can reduce the digestibility of semi-moisture feed with whey protein. However, semi moisture feed without whey protein may be provided to newly weaned piglets.

Key Words: growth performance, nutrient digestibility, whey protein, weaning pig.

P113 Meta-analysis of the effect of sodium butyrate supplementation on the performance and intestinal morphology of postweaning piglets. L. Arnalot, W. Lambert, and T. Chalvon-Demersay*, *METEX NØØVIST-AGO, Paris, France.*

Sodium butyrate supplementation is a nutritional approach to improve the barrier and digestive functions of the intestine in monogastrics. However, to date there is no meta-analysis summarizing the effects of this additive on the performance and intestinal morphology of postweaning piglets. A systematic search on Pubmed and Google Scholar combining the following keywords: [Piglet] OR [Pig] AND [Butyrate] OR [Butyric acid] AND [Performance] OR [Gut morphology] was performed and identified 11 studies investigating the effect of sodium butyrate supplementation on performance and/or intestinal morphology in postweaning piglets. Data were expressed as a percentage of unsupplemented treatment and analyzed with Minitab software using a general linear model with the study as a fixed factor and the dose of supplementation as a covariate. Over the first 2 weeks after weaning, sodium butyrate supplementation was associated with improved body weight gain (P = 0.004), feed intake (P = 0.010) and feed conversion (P = 0.002). Over the complete experimental period, the supplementation was associated with improved gain (P = 0.025) and a trend toward increased feed intake (P = 0.064). No significant effect of supplementation was observed on feed efficiency (P > 0.05) or on intestinal morphology parameters (P > 0.05). This study confirms the effectiveness of sodium butyrate supplementation to improve piglet performance, especially during the first days after weaning. This effect does not seem to be mediated by an improvement of gut morphology.

Key Words: piglets, performance, butyrate, gut health, meta-analysis

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