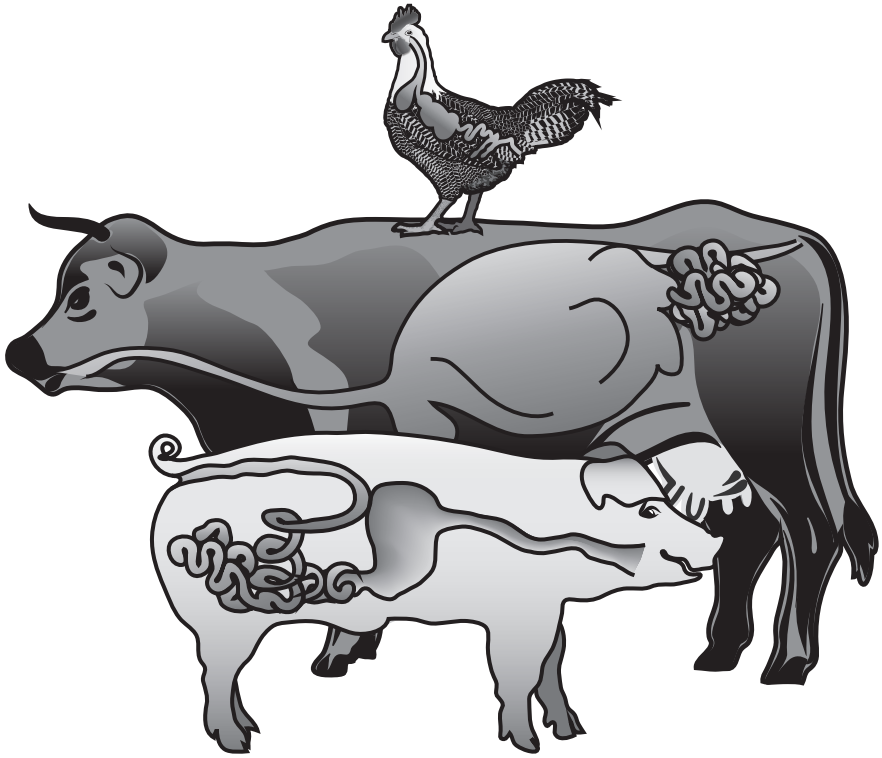
Symposium on Gut Health
in Production of Food Animals
November 9–11, 2015, Kansas City, Missouri



Program and Abstracts
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CONTENTS

WELCOME

On behalf of the Organizing Committee for the 4th Symposium on Gut Health in Production of Food Animals, I welcome you back to Kansas City, Missouri! After a very successful 2nd Symposium here at the Marriott Country Club Plaza in 2013, we decided a return trip was in order. I look forward to another scientifically and socially rewarding meeting in 2015.

Like the first three symposia organized around the topic of gut health in food animals, the aim 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 concept 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. Understanding the crosstalk between ALL of these interrelated components of the gut is what cumulatively makes the gut the basis for the well-being of animals and the motor that drives their performance. The research described in the abstracts from this symposium are elucidating these links and mechanisms that interconnect the three components of the gut and how each can be manipulated to improve animal health.

This year we have to invited four distinguished plenary speakers who will cover current research topics in avian, porcine, bovine, and, for the first time, fish gut health. Please take advantage of the presence of these scientists to engage in productive talks and develop collaborations between laboratories to further the science of gut health.

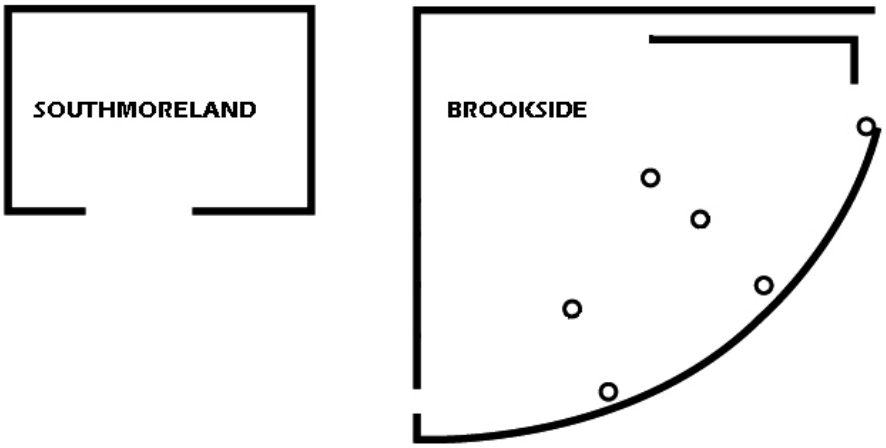
Likewise, I encourage all of you to benefit from informal nature of the symposium—it was planned this way to maximize interaction between scientists. I again ask that senior researchers make a special effort to engage with the graduate students who are attending and presenting. Remember that, whatever 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 Kansas City!

Mike Kogut
Chair, Organizing Committee

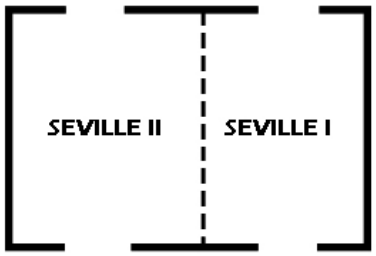
Marriott Kansas City Country Club Plaza

FIRST FLOOR

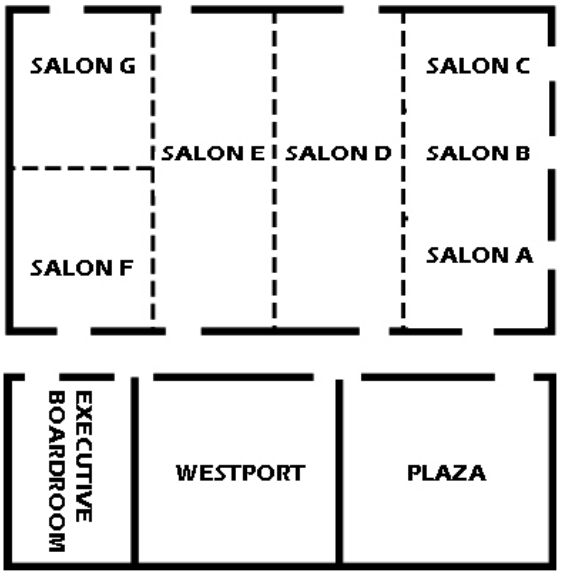


SECOND FLOOR

SEVILLE BALLROOM



GRAND BALLROOM



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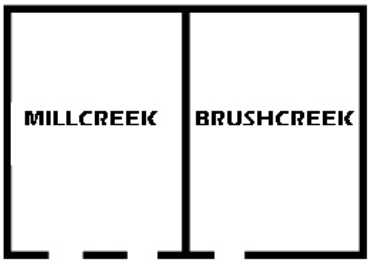

Program

Sunday, November 8

5:00 pm – 7:00 pm Registration: Grand Ballroom Foyer

Monday, November 9

7:00 am – 8:00 am Breakfast: Grand Ballroom EFG
Sponsored by King Techina Group

7:00 am – 5:00 pm Registration: Grand Ballroom Foyer

SESSION I

Chair: Mike Kogut, USDA-ARS
Grand Ballroom ABCD

8:15 am Welcome.
Mike Kogut, USDA-ARS, Chair, Organizing Committee.

8:30 am – 9:30 am Dietary modulation of intestinal epithelial defense in chickens. (Abstract 100)
G. Zhang, Department of Animal Science, Oklahoma State University, Stillwater, OK, USA.*

9:30 am – 10:00 am Degenerative primer design and gene sequencing validation for select turkey genes. (Abstract 101)
S. Loeffler, M. Lilburn, and M. Wick, The Ohio State University, Columbus, OH, USA.*

10:00 am – 10:30 am Effects of coated highly dispersed nano zinc oxide dietary supplements on growth performance and diarrhea symptoms in weaning piglets. (Abstract 102)
B. Zou, L. Zhe, R. Yu, and S. Li, King Techina Group.*

10:30 am – 11:00 am Coffee Break: Grand Ballroom EFG
Sponsored by Phibro Animal Health

11:00 am – 11:15 am Dietary available phosphorus and phytase levels can influence the pH of the upper intestine and histomorphological characteristics of the lower intestine in broilers. (Abstract 103)
L. Beeson[*1,3], C. Walk[2], P. Hastie[3], M. Bedford[2], and O. Olukosi[1], [1]Monogastric Science Research Centre, SRUC, Edinburgh, United Kingdom, [2]AB Vista, Marlborough, Wiltshire, United Kingdom, [3]University of Glasgow, Glasgow, United Kingdom.*

11:15 am – 11:45 am Microbiota characterization and functional potential of rumen fluid of high- and low-producing dairy cattle. (Abstract 104)
S. Lima[*1,2], R. Pereira[1], V. Machado[1], and R. Bicalho[1], [1]Cornell University, Ithaca, NY, USA, [2]Science Without Borders, Brazil.*

11:45 am – 1:00 pm Lunch (provided): Grand Ballroom EFG

1:00 pm – 3:00 pm Poster Session: Grand Ballroom EFG

SESSION II

Chair: Mike Kogut, USDA-ARS
Grand Ballroom ABCD

3:00 pm – 4:00 pm Understanding host-microbial interactions and their effect on gut development in newborn dairy calves. (Abstract 105)
N. Malmuthuge[1], G. Liang[1], P. Griebel[2], and L. L. Guan[*1], [1]University of Alberta, Edmonton, AB, Canada, [2]University of Saskatchewan, Saskatoon, SK, Canada.*

4:00 pm – 4:30 pm Influence of calcium source, dietary calcium level, and probiotic on naturally occurring necrotic enteritis in broilers. (Abstract 106)
A. McElroy[*1] and D. Paiva[2], [1]Texas A&M University, [2]FDA.*

4:30 pm – 5:00 pm Long-term benefits of modifying pigs feed presentation on *Salmonella* species shedding, digestive microbiota, and health. (Abstract 107)
P. LeBel[*1], A. Letellier[1], B. Laplante[2], E. Yergeau[3], and P. Fravalo[1], [1]Research Chair in Meat Safety, Faculty of Veterinary Medicine, University of Montreal, St-Hyacinthe, QQ, Canada, [2]F. Ménard, L'Ange-Gardien, QC, Canada, [3]National Research Council Canada, Energy, Mining and Environment, Montreal, QC, Canada.*

6:00 pm – 8:00 pm Reception: Grand Ballroom EFG
Sponsored by Merck Animal Health

Tuesday, November 10

7:00 am – 8:00 am Breakfast: Grand Ballroom EFG
Sponsored by Arm and Hammer Animal Nutrition

7:00 am – 4:30 pm Registration: Grand Ballroom Foyer

SESSION III

Chair: Mike Kogut, USDA-ARS
Grand Ballroom ABCD

8:30 am – 9:30 am *Salmonella* in food animal production: Defining the need for antibiotic alternatives and exploring approaches for intervention. (Abstract 108)
S. Bearson, USDA, ARS, National Animal Disease Center.*

9:30 am – 10:00 am Evolution of the fecal microbiome of the sow during the gestation and its effect on *Salmonella* excretion. (Abstract 109)
G. Larivière-Gauthier[*1], A. Letellier[1], É Yergeau[2], B. Laplante[3], and P. Fravalo[1], [1]Research Chair in Meat Safety, Université de Montréal, Saint-Hyacinthe, Québec, Canada, [2]National Research Council Canada, Energy, Mining and Environment, Montréal, Québec, Canada, [3]F. Ménard Inc, L'Ange-Gardien, Québec, Canada.*

10:00 am – 10:30 am Infection of broilers with *Eimeria* causes species-specific changes in mRNA expression of genes associated with amino acid and sugar uptake in the gut. (Abstract 110)
K. B. Miska and R. H. Fetterer, USDA/ARS, Beltsville, MD, USA.*

10:30 am – 11:00 am Coffee Break: Grand Ballroom EFG
Sponsored by Purina

11:00 am – 11:30 am Bacteremia and lameness: An investigation into blood microbiota associated with bacterial chondronecrosis with osteomyelitis (BCO) in broilers. (Abstract 111)
R. K. Mandal[*1], T. Jiang[1], R. F. Wideman[1], A. A. Alrubaye[2], D. D. Rhoads[2], I. Pevzner[3], and Y. M. Kwon[1,2], [1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]Cell and Molecular Biology Program, University of Arkansas, Fayetteville, AR, USA, [3]Cobb-Vantress Inc, Siloam Springs, AR, USA.*

11:30 am – 12:00 pm Effects of virginiamycin and (or) a phytogenic feed additive on performance and intestinal health of broiler chickens. (Abstract 112)
H. M. Cervantes[*1,2], M. J. Da Costa[2], K. W. Bafundo[1], and G. M. Pesti[2], [1]Phibro Animal Health, [2]University of Georgia.*

12:00 pm – 2:00 pm Lunch (provided): Grand Ballroom EFG
Sponsored by Cobb

SESSION IV

Chair: Mike Kogut, USDA-ARS
Grand Ballroom ABCD

2:00 pm – 3:00 pm Toward an understanding of fish gut tolerance to alternative feeds—The Trout-Grains Project. (Abstract 113)
J. Abernathy[*1], A. Brezas[2], T. Welker[1], K. Liu[3], F. Barrows[4], K. Overturf[1], and R. Hardy[2], [1]USDA-ARS, Hagerman Fish Culture Experiment Station, [3059]F National Fish Hatchery Rd, Hagerman, ID, USA, [2]University of Idaho, Hagerman Fish Culture Experiment Station, Hagerman, ID, USA, [3]USDA-ARS, Small Grains and Potato Germplasm Research Unit, Aberdeen, ID, USA, [4]USDA-ARS, Bozeman Fish Technology Center, Bozeman, MT, USA.*

3:00 pm – 3:30 pm Use of a novel enzymatic strategy (Hemicell-L) to assess the effect on broiler intestinal health and productivity under commercial conditions in a German integration. (Abstract 114)
M. A. Martinez-Cummer[*1], G. González-García[2], and K. Poulsen[2], [1]Elanco Animal Health, Greenfield, IN, USA, [2]S.A. Eli Lilly Benelux N.V, Antwerp, Belgium.*

3:30 pm – 4:00 pm Combining feed enzyme activities in poultry feeds: Adding energy-sparing enzymes to manage the effect of unnecessary feed-induced immune responses on broiler intestinal health and overall production performance. (Abstract 115)
M. A. Martínez-Cummer, Elanco Animal Health, Greenfield, IN, USA.*

4:00 pm – 4:30 pm Effects of grain processing on starch digestion and performance of feedlot cattle. (Abstract 116)
E. F. Schwandt[*1], J. J. Wagner[2], T. E. Engle[2], S. J. Bartle[1], D. U. Thomson[1], and C. D. Reinhardt[1], [1]Kansas State University, Manhattan, KS, USA, [2]Colorado State University, Fort Collins, CO, USA.*

6:00 pm – 8:00 pm Reception: Grand Ballroom EFG
Sponsored by Merck Animal Health

Wednesday, November 11

7:00 am – 8:00 am Breakfast: Grand Ballroom EFG
Sponsored by Phileo

8:00 am – 11:00 am Registration: Grand Ballroom Foyer

SESSION V

Chair: Mike Kogut, USDA-ARS
Grand Ballroom ABCD

9:00 am – 9:30 am Effect of organic acids and essential oils on enteric mucosa morphology and histology as indicators of gut health in poultry. (Abstract 117)
R. D. Malheiros, V. M. B. Moraes, and P. R. Ferket, Prestage Department of Poultry Science, NC State University, Raleigh, NC, USA.*

9:30 am – 10:00 am Leaky gut and mycotoxins: Aflatoxin B_1 does not increase gut permeability in broiler chickens. (Abstract 118)
R. Galarza-Seeber, J. D. Latorre, A. Wolfenden, B. Hargis, and G. Tellez, University of Arkansas, Fayetteville, AR, USA.*

10:00 am – 10:30 am Life cycle of probiotic *Bacillus* in the gastrointestinal tract. (Abstract 119)
M. Bernardeau[1,2], M. J. Lehtinen[3], S. D. Forssten[3], and P. Nurminen[3], [1]DuPont-Danisco, Industrial Biosciences, Animal Nutrition, Marlborough, United Kingdom, [2]QAIEA research Unit, University of Caen, Caen, France, [3]DuPont Nutrition and Health, Kantvik, Finland.*

10:30 am – 11:00 am Effect of dietary supplementation of β-galacto-oligosaccharides on intestinal microarchitecture of broilers reared under cyclic heat stress. (Abstract 120)
S. Ashraf, H. Zaneb, H. Rehman, S. Muti, and S. Ijaz, University of Veterinary and Animal Sciences Lahore, Lahore, Punjab, Pakistan.*

Poster Presentations

P100 Daily feeding of *Lactobacillus animalis* improved performance and reduced mortality and morbidity associated with necrotic enteritis in broilers in two different model systems.
S. Lerner[*1], J. McNaughton[2], and G. Mathis[3], [1]Nutrition Physiology Company LLC, Kansas City, MO, USA, [2]AHPharma, Salisbury, MD, USA, [3]Southern Poultry Research, Athens, GA, USA.*

P101 Effect of a *Bacillus* direct-fed microbial candidate on nutrient digestibility, gut permeability, bone quality, and growth performance in broilers consuming a sorghum-based diet.
J. D. Latorre[*1], R. E. Wolfenden[2], J. L. Vicente[2], A. D. Wolfenden[1], R. Galarza-Seeber[1], V. A. Kuttappan[1], L. R. Bielke[1], B. M. Hargis[1], and G. Tellez[1], [1]University of Arkansas, Department of Poultry Science, Fayetteville, Arkansas, USA, [2]Pacific Vet Group, Fayetteville, Arkansas, USA.*

P102 Evaluation of a *Bacillus* spp. direct-fed microbial candidate for aflatoxin B_1 biodegradation in broiler chickens.
R. Galarza-Seeber, J. D. Latorre, A. Wolfenden, B. Hargis, and G. Tellez, University of Arkansas, Fayetteville, AR, USA.*

P103 Involvement of microRNAs in calf intestinal growth and development and probable modulatory role of *Saccharomyces cerevisiae*.
E. M. Ibeagha-Awemu, P.-L. Dudemaine, J. Chiquette, G. Talbot, and N. Bissonnette, Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, Quebec, Canada.*

P104 Sugar cane bagasse affects the fungal community dynamic in the sheep rumen.
E. M. Romagnoli[*1], C. Dunlap[2], A. L. Abdalla[3], and R. Mendes[1], [1]Embrapa Environment, Laboratory of Environmental Microbiolog, Jaguariuna, Sao Paulo, Brazil, [2]United States Department of Agriculture, Agricultural Research Service, National Center for Agricultural Utilization Research, Peoria, IL, USA, [3]University of Sao Paulo, Center for Nuclear Energy in Agriculture, Piracicaba SP, Brazil.*

P105 A comparison of fungal and bacterial populations in broilers from high- and low-producing farms.
J. Byrd, USDA, ARS, Food and Feed Safety Research Unit, College Station, TX, USA.*

P106 Genetic relatedness between avian pathogenic *Escherichia coli* recovered from broiler breeders and chicks reveals potential for vertical transmission.
J. Lambrecht[*1], F. Delago[1], K. Gibbs[2], T. Horne[3], and E. Galbraith[1], [1]DuPont Nutrition and Health, Waukesha, WI, USA, [2]DuPont Industrial Biosciences, Marlborough, United Kingdom, [3]Chemuniqué, Randburg, South Africa.*

P107 Investigation of avian pathogenic *Escherichia coli* (APEC) virulence-associated genes from US poultry isolates.
F. Delago, C. Urbain, J. Lambrecht, S. Gebert, and E. Galbraith, DuPont, Waukesha, WI, USA.*

P108 Effect of organic selenium on the chicken cecal microbiome equilibrium and *Campylobacter jejuni* carriage.
A. Thibodeau, P. Fravalo, and A. Letellier, Université de Montréal, Saint-Hyacinthe, Québec, Canada.*

P109 Metagenomic analysis of the bovine hindgut from *Salmonella* Kentucky and Cerro-shedding dairy cows.
B. Haley, J. Karns, and J. Van Kessel, Environmental Microbial and Food Safety Laboratory, Beltsville Agricultural Research Center, Agricultural Research Service, US Department of Agriculture, Beltsville, MD, USA.*

P110 Effect of stress events on mucosal permeability, bacterial translocation, and lameness in broilers.
L. R. Bielke[*1], J. C. Bielke[1], V. A. Kuttappan[2], E. A. Vicuña[2], A. Al-Ogaili[2], J. D. Latorre[2], B. D. Mahaffey[2], B. M. Hargis[2], and G. I. Tellez[2], [1]Ohio State University, Department of Animal Sciences OARDC, Columbus, OH, USA, [2]University of Arkansas, Poultry Science Department Poultry Health Laboratory, Fayetteville, AR, USA.*

P111 The effect of sericea lespedeza on coccidia-infected chickens.
L. Trinh, Louisiana State University, Baton Rouge, LA, USA.*

P112 Effect of seasonality in the intestinal integrity of broilers in Northern European countries in the period 2012–2014: Results of the Elanco Health Tracking System.
A. Zocche and G. Garcia, Elanco Animal Health, Greenfield, IN, USA.*

P113 Systems kinomics reveal that *Salmonella enterica* Enteritidis modulates host immune signaling pathways in the cecum of chickens that are associated with the establishment of persistent infections.
M. H. Kogut[*1], C. L. Swaggerty[1], R. Salvaraj[2], and R. J. Arsenault[3], [1]USDA-ARS, College Station, TX, USA, [2]Ohio State University, Wooster, OH, USA, [3]University of Delaware, Newark, DE, USA.*

P114 Comparison of immunometabolic response in turkeys orally inoculated with *Salmonella* Heidelberg.
R. J. Arsenault[*1], M. H. Kogut[2], H. He[2], and K. Genovese[2], [1]University of Delaware, Department of Animal and Food Sciences, Newark, DE, USA, [2]United States Department of Agriculture, College Station, TX, USA.*

P115 Effects of supplementation of probiotics (*Bacillus*, *Lactobacillus*, *Aspergillus niger*) and prebiotics (chicory, rice bran) on growth performance, nutrient digestibility, meat quality, excreta microbiota, hematological profile, and excreta noxious gas emissions in broilers.
W. Yun, H. S. Choi, H. G. Choi, Y. I. Choi, and J. H. Cho, Chungbuk National University, Cheongju, Chungbuk, South Korea.*

P116 Effects of direct-fed microbials on growth performance, gut microbiota, and pathogen resistance in *Litopenaeus vannamei* shrimp.
J. Barnes[*1], J. P. Gorsuch[1], M. S. Showell[1], C. L. Kitts[2], A. L. Lawrence[3], and R. S. Carpenter[1], [1]BiOWiSH Technologies Inc, Cincinnati, OH, USA, [2]California Polytechnic State University, San Luis Obispo, CA, USA, [3]Ralco Animal Nutrition, Marshall, MN, USA.*

P117 The effect of intra-amniotic and post-hatch dietary synbiotic administration on duodenum *MUC2* gene expression.
A. Calik[*1], F. Dilber[1], P. Sacakli[1], and T. Tekinay[2], [1]Department of Animal Nutrition Nutritional Diseases, Faculty of Veterinary Medicine, Ankara University, Ankara, Turkey, [2]Life Sciences Research and Application Center, Gazi University, Ankara, Turkey.*

P118 Effect of garlic powder and vitamin E-selenium and their combination on performance, immune response, lipid profile, and blood picture of broilers.
Y. J. Jameel[*1,2], M. Hashim[2], M. A. Husain[1], and A. M. Sahib[3], [1]University of Kerbala, Karbala, Iraq, [2]Texas A&M University, College Station, TX, [3]University of Kufa, Kufa, Najaf, Iraq.*

P119 Effects of suckling pigs from gilts fed dried bovine plasma during gestation and lactation on intestinal morphometry.
B. V. Freitas[*1], R. A. Nacimento[1], S. M. M. K. Martins[1], L. F. Araújo[1], C. S. S. Araújo[1], and G. Hosotani[2], [1]University of São Paulo, Pirassununga, SP, Brazil, [2]University of Missouri, Columbia, MO, USA.*

P120 Effect of monocomponent protease on performance, organ size, and duodenal morphology in broilers fed autoclaved soybean.
M. E. Mayorga[*1] and L. B. Moraes[2], [1]UNIAGRARIA, Bogota, Cundinamarca, Colombia, [2]Institute for Veterinary Research "Desidério Finamor" (IPVDF), Animal Health, State Foundation of Agricultural Research (Fepagro), Eldorado do Sul, Rio Grande do Sul, Brazil.*

NOTES

Oral Presentations
Session I

100 Dietary modulation of intestinal epithelial defense in chickens.

G. Zhang*,
Department of Animal Science, Oklahoma State University, Stillwater, OK, USA.

Subtherapeutic use of antibiotics in livestock production for growth promotion and disease prevention has the potential to drive up antimicrobial resistance. Although various forms of alternatives to antibiotics have been explored, none is able to match the efficacy of antibiotics. Host defense peptides (HDP) constitute a diverse group of small molecules that are an important component of innate immunity with potent antimicrobial, immunomodulatory, and barrier function-enhancing activities. We recently discovered several classes of small-molecule compounds that are highly efficient in specific induction of endogenous HDP. Supplementation of these HDP-inducing compounds enhances bacterial clearance and intestinal barrier integrity with a tendency to improve animal production efficiency as well. Unlike most immunomodulators on the market, these HDP-inducing compounds generally have a minimal effect on the inflammatory response and therefore have potential for further development as antibiotic alternatives for disease control and prevention in animal agriculture. In the presentation, I will highlight some of the newly identified dietary compounds and their efficacy in promoting growth, enteric defense, and barrier function in the context of gut inflammation in broilers.

Key words: host defense peptides, antimicrobial peptides, epithelial defense

101 Degenerative primer design and gene sequencing validation for select turkey genes.

S. Loeffler*, M. Lilburn, and M. Wick,
The Ohio State University, Columbus, OH, USA.

We successfully designed and validated degenerative primers for turkey genes *MUC2*, *RPS13*, *TBP*, and *TFF2* based on chicken sequences. Analyzing gene transcription differences in response to treatments has been established as a way to evaluate an animal's physiological response to changes in its environment. This particular technique requires known gene sequences to develop primers to amplify specific genes of interest using PCR. This poses a problem when working with the turkey, as few genes have been sequenced. To use gene transcription analysis to evaluate the mucin response to probiotic supplementation in turkeys, we used multiple techniques including degenerate sequence analysis to determine gene-specific primers. Primers were designed for the genes *MUC2*, *TFF2*, *RPS13*, and *TBP* using a degenerative primer design method based on the available *Gallus gallus* sequences. All primer sets that produced a single PCR amplicon of the expected size were cloned into the TOPO vector and then transformed into TOP10 competent cells. Plasmid DNA isolation was performed on the TOP10 cell culture and sent for sequencing. Sequences were then analyzed for homology using the NCBI BLAST tool. All genes sequenced had over 90% homology with both the chicken and predicted turkey sequences. The sequences were used to design new homologous primer sets for the genes of interest.

102 Effects of coated highly dispersed nano zinc oxide dietary supplements on growth performance and diarrhea symptoms in weaning piglets.

B. Zou, L. Zhe, R. Yu*, and S. Li,
King Techina Group.

High dosage of zinc oxide in the diet has been a major method of controlling diarrhea in weaning piglets. However, continuous use results in severe negative effects on the environment and growth performance of the pigs in later periods. Research indicates that highly dispersed materials have smaller particle size and larger surface area, so the addition of highly dispersed zinc oxide can reduce the dosage of zinc oxide. However, highly dispersed nano zinc oxide reacts easily with gastric acid in the stomach to form zinc chloride, which reduces the antidiarrheal effect. In this study, we treated nano zinc oxide with a target-release coating to study the effect of coated highly dispersed nano zinc oxide (CNZ) dietary supplements on the growth performance and diarrhea symptoms in weaning piglets. The experiment used 120 healthy weaning piglets similar in age and weight (DLY, n = 120, 8.35 ± 0.01 kg of BW), randomly assigned to 4 groups of different treatments, each with 3 repeats of 10 piglets. Group 1 (PZ) was the control group, fed a basal diet with 2,450 ppm of powdery zinc oxide. Group 2 (CNZ50), group 3 (CNZ75), and group 4 (CNZ100) were experimental groups fed basal diets with coated highly dispersed nano zinc oxide in the amounts of 50, 75, and 100 ppm, respectively. The experimental lasted 44 d. There were no significant differences in growth performance among the 4 groups ($P > 0.05$). Compared with the PZ control group, the feed:gain ratio of the CNZ50, CNZ75, and CNZ100 experimental groups decreased, respectively, by 3.66, 3.14, and 1.57%, but their daily weight gain and feed intake were not significantly different. The diarrhea incidence of CNZ100 and PZ groups were not significantly different, whereas diarrhea incidence in theCNZ50 and CNZ75 groups were significantly higher than that of the PZ group ($P < 0.01$). Among CNZ groups, the diarrhea incidence and index of the piglets decreased as the level of CNZ increased. The diarrhea index of the PZ group was significantly ($P < 0.05$) lower than the other groups. The results show that, in this experiment, CNZ and PZ yield similar results in promoting growth, and the optimal dose of CNZ for controlling diarrhea was 100 ppm.

103 Dietary available phosphorus and phytase levels can influence the pH of the upper intestine and histomorphological characteristics of the lower intestine in broilers.

L. Beeson*[1,3], C. Walk[2], P. Hastie[3], M. Bedford[2], and O. Olukosi[1],
[1]Monogastric Science Research Centre, SRUC, Edinburgh, United Kingdom, [2]AB Vista, Marlborough, Wiltshire, United Kingdom, [3]University of Glasgow, Glasgow, United Kingdom.

Three hundred eighty-four Ross 308 broilers were allocated to 6 dietary treatments for 22 d. Diets were a nutritionally adequate positive control (PC; adequate available phosphorus, aP) and negative control diet (NC; marginally deficient in aP and Ca, relative to PC) supplemented with 0, 500 or 1,500 FTU/kg phytase, in a 2 × 3 factorial arrangement. Following euthanasia by overdose of barbiturate, 2 birds per pen were used to measure indices of gut morphology. pH was taken in the duodenum (DUO), jejunum (JEJ), ileum (ILE), and cecum (CAE) and sections of DUO, JEJ, and ILE were collected for histomorphology. Data were analyzed using Genstat; significance was determined at $P \leq 0.05$. Birds fed the PC diets tended to have an increase in DUO pH ($P = 0.054$). There was an aP × phytase interaction ($P < 0.01$) for JEJ pH, with phytase increasing pH in the PC (quadratic, $P = 0.001$) and NC (linear, $P < 0.01$). In the DUO, there was aP × phytase interaction ($P < 0.05$) on apparent villi surface area (SA); phytase increased SA in the NC (quadratic, $P < 0.05$), being greatest with 500 FTU, but requiring 1500 FTU in the PC. There was a quadratic increase in basal width as phytase increased ($P < 0.05$). In the JEJ, there was aP × phytase interaction ($P < 0.05$), with apical width increasing (linear, $P < 0.01$) in the NC with phytase but there was no effect of phytase in birds fed the PC. There was a tendency for aP × phytase interaction ($P = 0.09$) on crypt depth (Cd), whereas phytase increased Cd in the NC (quadratic, $P = 0.09$), but not the PC. There were interactive aP × phytase ($P < 0.05$) effects on ILE villi number and Cd; phytase in the PC increased villi numbers (quadratic, $P < 0.05$) and reduced Cd (linear, $P < 0.05$); however, these effects were not seen in the NC. Birds fed the PC had more crypts than birds fed the NC ($P < 0.05$). Phytase and aP appear to influence pH in the proximal intestine and intestinal morphology. This may be associated with phytate destruction and reduced endogenous cell sloughing in the proximal intestine and result in increased nutrient utilization, particularly in birds fed low aP diets.

104 Microbiota characterization and functional potential of rumen fluid of high- and low-producing dairy cattle.

S. Lima*[1,2], R. Pereira[1], V. Machado[1], and R. Bicalho[1],
[1]Cornell University, Ithaca, NY, USA, [2]Science Without Borders, Brazil.

The objective of this study was to characterize the ruminal fluid microbiota and its functional potential in high- and low-producing dairy cattle using shotgun metagenomic sequencing. A total of 8 ruminal fluid samples were collected according to milk production status: 4 from animals labeled as high-milk-production cows (HPC), and 4 from animals labeled as low-milk-production cows (LPC). Regardless of milk production label of cows, the ruminal fluid had a diverse microbial community and included Bacteria, Eukarya, Archaea, and Virus. The most common phyla in the Bacteria domain were Firmicutes, Bacteroidetes, Proteobacteria, and Actinobacteria. The top 5 bacteria genera of ruminal fluid of both HPC and LPC were *Prevotella*, *Clostridium*, *Bacterioides*, *Ruminococcus*, and *Eubacterium*. The HPC had a higher mean number of hits of *Paramecium tetraurelia* as well as *Tetrahymena thermophile* (members of Eukarya domain) compared with LPC. The Archaea community of HPC and LPC was mostly composed of methanogen organisms. Select viruses were unique to cattle with different milk production labels, with 19 phages present only in the ruminal fluid of HPC and 16 phages present only in the ruminal fluid of LPC. The HPC ruminal fluid microbiota had a significantly higher mean number of hits for protein biosynthesis, protein processing and modification, protein degradation, protein folding, and selenoprotein compared with LPC ruminal fluid microbiota. Fluoroquinolone resistance, cobalt-zinc-cadmium resistance, and multidrug resistance efflux pumps and methicillin resistance in *Staphylococcus* were highly abundant in all ruminal fluid microbiotas tested. This study provides a deep insight into rumen microbial community and its dynamics. In conclusion, the ruminal fluid virome differed between HPC and LPC and ruminal fluid microbiota from HPC had a higher abundance of genes associated with protein predicted metabolisms than LPC.

Session II

105 Understanding host-microbial interactions and their effect on gut development in newborn dairy calves.

N. Malmuthuge[1], G. Liang[1], P. Griebel[2], and L. L. Guan*[1],
[1]University of Alberta, Edmonton, AB, Canada, [2]University of Saskatchewan, Saskatoon, SK, Canada.

Establishment of a host-specific gut microbiota plays a crucial role in development of the mucosal immune system in newborn mammals and influences their susceptibility to enteric infections. Using functional genomics-based approaches, we explored host-microbial interactions in the neonatal bovine gut by analyzing microbial colonization, its influence on the host, and the effect of external factors such as diet. Colonization of the calf gastrointestinal tract (GIT) began during birthing and bacterial density reached 10^8 16S rRNA gene copy/g of GIT (contents and tissue) within 30 min of birth. Bacterial density increased 100-fold within the first 12 h of life, only when calves were fed colostrum soon after birth, but not when colostrum was withheld for 6 or 12 h postpartum. This observation confirms that timed feeding of colostrum affects microbiome development. Approximately 10% of jejunal and ileal bacteria were *Bifidobacterium*, a beneficial bacterium, which was dominated by *B. longum infantis* within 30 min after birth. The feeding of heat-treated colostrum soon after birth increased small intestinal mucosa-attached *Bifidobacterium* by 3.2-fold and 5.2-fold compared with fresh colostrum and no colostrum calves, respectively, at 6 h after birth. The prevalence of *Bifidobacterium* gradually increased during the first 3 wk of life and then declined by 6 wk of age. It was also correlated with the expression of immunoregulatory genes (IL8, IL10) and microRNAs (miR15/16, 196, 29) involved in regulating T-cell and dendritic cell development. These correlations suggest a link between microbiome colonization and the regulation of neonatal mucosal immune system development and function. Our studies reveal that microbiome colonization of the bovine GIT begins during birthing and the abundance of beneficial bacteria, such as *Bifidobacterium*, is altered by colostrum feeding. The prevalence of *Bifidobacterium* then has a significant effect on subsequent development of the mucosal immune system.

106 Influence of calcium source, dietary calcium level, and probiotic on naturally occurring necrotic enteritis in broilers.

A. McElroy*[1] and D. Paiva[2],
[1]Texas A&M University, [2]FDA.

Clostridium perfringens (CP), the bacteria responsible for necrotic enteritis (NE), are naturally occurring in the intestines of chickens; however, their presence alone is not a determining factor for disease development. Predisposing factors leading to an overgrowth of CP are crucial to NE development and have been reported to include dietary factors such as protein level and source, cereal type, and feed form. Little research has evaluated the role of dietary mineral levels or exogenous feed enzymes during natural occurrence of NE and how dietary alterations can be combined with probiotic supplementation to support intestinal integrity. Our laboratory performed several trials evaluating the influence of dietary Ca (level and source) and dietary probiotic inclusion on broiler performance, mineral digestibility, bone ash, and intestinal morphology during naturally occurring NE. Broilers were reared on dirty litter from flocks experiencing NE, and dietary treatments included multiple Ca sources (limestone or highly soluble calcified seaweed), 0.6 or 0.9% dietary Ca, and dietary probiotic inclusion. Significance was determined at $P < 0.05$. Mortality from NE was significantly increased with inclusion of 0.9% dietary Ca or 1,000 FTU/kg phytase. Additionally, an interaction between Ca source and Ca levels occurred, with higher mortality when broilers were fed 0.90% calcified seaweed diets compared with 0.60% Ca diets (regardless of Ca source) and 0.90% Ca diets formulated with limestone. At higher levels of Ca inclusion, probiotic inclusion provided no benefit to bird performance or mortality from NE. However, reducing the dietary level of Ca to 0.6% resulted in reduced mortality from NE, and with the inclusion of probiotic, resulted in heavier BW. The results suggest that dietary Ca level and source affect NE pathogenesis and associated mortality, which can be combined with probiotic supplementation for beneficial effects on broiler intestinal health.

107 Long-term benefits of modifying pigs feed presentation on *Salmonella* species shedding, digestive microbiota, and health.

P. LeBel*[1], A. Letellier[1], B. Laplante[2], E. Yergeau[3], and P. Fravalo[1],
[1]Research Chair in Meat Safety, Faculty of Veterinary Medicine, University of Montreal, St-Hyacinthe, QQ, Canada, [2]F. Ménard, L'Ange-Gardien, QC, Canada, [3]National Research Council Canada, Energy, Mining and Environment, Montreal, QC, Canada.

Salmonellosis is one of the most important foodborne illness in Canada. According to many epidemiological studies, a promising on-farm intervention is the use of mash feed instead of the typically used pellet feed. However, these studies failed to clearly isolate this variable between the compared groups. Moreover, the feed change has been associated with a lower feed conversion and the benefit on *Salmonella* is often only serological. In this study, over 900 pigs in the same building were assigned a diet varying only in their processing and particle size, during the entire finishing period, leading to 4 experimental groups: pellet with small particle (PS), pellet with large particle (PL), mash with small particle (MS), or mash with large particle (ML). Individual fecal samples were taken at the farm 2 wk before slaughter, and intestinal content and ileal wall on 24 pigs per group were sampled at slaughter. At the end of the finishing period, significantly more pigs (9/24) shed *Salmonella* in the group fed the industrial reference feed (PS) than in the 3 other experimental groups (MS = 0/24; ML = 2/24; PL = 2/24). Also, a global *Salmonella* shedding benefit from the use of a mash feed compared with a pellet feed regardless of the particle size was observed on the farm (11/48 vs. 2/48) and in the rectal content at slaughter (17/48 vs. 7/48). Real-time PCR quantification showed significantly more *Bifidobacterium* spp. in pig feces on the farm when mash and large particle size diets were used and a significantly greater ratio of lactobacilli/enterobacteria

in the feces on the farm and in the rectal content when a large particle size feed was used. Next-generation sequencing is being done on the same samples to extend the digestive flora analysis. Histological analysis on ileal wall showed a tendency to a greater proportion of acid mucin-producing goblet cells when a mash feed was used, and more samples are being analyzed to deepen our analysis. A similar feed conversion rate was obtained with the use of PS and PL, and because the latter provides gut health and a *Salmonella* spp. benefit, it might be the key to a cost-efficient solution for producers.

Session III

108 *Salmonella* in food animal production: Defining the need for antibiotic alternatives and exploring approaches for intervention.

S. Bearson*,
USDA, ARS, National Animal Disease Center.

Salmonella continues to be one of the most common causes of bacterial foodborne infections in the United States. Listed as a Serious Threat Level pathogen by the Centers for Disease Control and Prevention, multidrug-resistant (MDR) *Salmonella* leads to higher rates of morbidity (humans) and mortality (livestock) compared with antibiotic-sensitive strains. Antibiotics are given to animals and humans to control bacterial diseases. Unfortunately, antibiotic administration can have unintended consequences for the bacteria colonizing a host, such as selection for antibiotic resistance, enhanced pathogen virulence, and induction of horizontal gene transfer. Investigations by our group have shown that exposure of certain MDR *Salmonella* to antibiotics important in the treatment of human and veterinary diseases will accelerate the *Salmonella* invasion response as well as induce phage-mediated transfer of virulence and antibiotic resistance genes. Our goal is to identify antibiotics that stimulate bacterial virulence mechanisms (as well as those that do not) and characterize the effects those antibiotics have on MDR *Salmonella*. This information will assist veterinarians and food animal producers when determining the proper antibiotic therapy for infectious disease treatment that does not unintentionally complicate an illness and compromise animal management. As antibiotic usage in animal production becomes increasingly scrutinized, a second goal of our research program addresses the need for alternatives to antibiotics in food animals. Multiple targeted interventions against *Salmonella* are under investigation by our research team, including a rationally designed DIVA vaccine, research-identified probiotic strains, and biotherapeutics such as immunomodulators. Identification of novel mechanisms in the relationship between antibiotic usage, MDR *Salmonella*, the host immune response, and the gastrointestinal microbiota will facilitate the improvement and development of intervention strategies directed against *Salmonella* on the farm.

109 Evolution of the fecal microbiome of the sow during the gestation and its effect on *Salmonella* excretion.

G. Larivière-Gauthier*[1], A. Letellier[1], É Yergeau[2], B. Laplante[3], and P. Fravalo[1],
[1]Research Chair in Meat Safety, Université de Montréal, Saint-Hyacinthe, Québec, Canada, [2]National Research Council Canada, Energy, Mining and Environment, Montréal, Québec, Canada, [3]F. Ménard Inc., L'Ange-Gardien, Québec, Canada.

Pork meat is estimated to be responsible for 10 to 20% of human salmonellosis cases. Control strategies on the farm could help to reduce contamination at the slaughterhouse. One of the targeted sectors of production is the maternity pen, where sows could be *Salmonella* reservoirs. The aim of this study was to characterize the fecal microbiome of sows excreting or not *Salmonella* during gestation phases. A total of 76 sows were selected and fecal matter was analyzed at the beginning or the end of gestation and *Salmonella* detection was conducted. Among the 76 sows tested, 31 were shedding *Salmonella*. The sows in the first third of their gestation shed significantly more frequently *Salmonella* (22/29) than those in the last third (9/47) (χ^2: $P < 0.05$). The shedding status of 19 of the sows that were previously sampled in the first third of their gestation was followed, this time in the last third, confirming reduction of the shedding. The association between changes in the intestinal microbiome during time and the evolution of *Salmonella* shedding has been explored. MiSeq sequencing has been conducted on the feces of 46 sows to identify shifts in the composition or diversity in the microbial community that could be associated with these variations. The v3–4 region of the 16S rRNA gene of the bacterial and archaeal populations was targeted. A mean of 1.23×10^5 sequences per sample was obtained and 289 bacterial and 15 archaeal taxon were represented. Significant changes in the composition of the microbiota were detected between sow at the beginning and the end of gestation (weighted and unweighted unifrac: $P < 0.05$) but also between *Salmonella* positive and negative sows (weighted unifrac: $P < 0.05$). On the taxonomic level, 3 populations (*Rothia*, Clostridium XIVa, Planctomycetia) were significantly associated with the beginning of the gestation and the presence of *Salmonella*, and 1 population (*Ruminococcus*) was associated with the end of the gestation and the absence of *Salmonella*. Thus, it would be interesting to further investigate the effect of stimulation of the *Ruminococcus* genus in the fight against *Salmonella*.

110 Infection of broilers with *Eimeria* causes species-specific changes in mRNA expression of genes associated with amino acid and sugar uptake in the gut.

K. B. Miska* and R. H. Fetterer,
USDA/ARS, Beltsville, MD, USA.

Coccidiosis in chickens is caused by infection of gut epithelial cells with protozoan parasites of the genus *Eimeria*. This disease causes losses to the poultry industry because infected birds fail to gain weight as rapidly as noninfected birds. For the present study, we determined the effect of *Eimeria* on expression of components of amino acid and sugar uptake mechanisms. Ross broilers were infected with *Eimeria maxima*, which infects the jejunum, *Eimeria acervulina*, which infects the duodenum, or *Eimeria tenella*, which infects the ceca. Sections of the jejunum, duodenum, and ceca (depending on species of *Eimeria*) were taken at several time points between d 0 and 14 post-infection (PI) for mRNA expression analysis. Genes examined included 1 digestive enzyme, 7 peptide and amino acid transporters located on the brush border, 8 transporters located on the basolateral surface of the gut epithelium, and 5 sugar transporters. All 3 *Eimeria* species examined caused decrease in expression of brush border transporters, particularly at d 5 to 7 PI, when pathology is greatest. The same pattern was seen in expression of sugar transporters. However, the expression of basolateral transporters differed among species. *Eimeria tenella* infection resulted in decreased expression of all basolateral transporters, whereas *E. maxima* infection caused increased expression of 2 genes and slight decrease in expression of the remaining 5 genes. Infection with *E. acervulina* resulted in increased expression at height of infection (5 d PI) of all brush border transporters. In conclusion,

Eimeria infection causes a general decrease in gene expression of sugar transporter and brush border amino acid transporters at the height of infection. However, the expression of basolateral transporters is increased in *E. maxima*- and *E. acervulina*-infected birds. It is possible that decreased expression of brush border transporters in combination with increased expression of basolateral transporter leads to decrease of nutrients available for the parasite, thus limiting parasite reproduction.

111 Bacteremia and lameness: An investigation into blood microbiota associated with bacterial chondronecrosis with osteomyelitis (BCO) in broilers.

R. K. Mandal*[1], T. Jiang[1], R. F. Wideman[1], A. A. Alrubaye[2], D. D. Rhoads[2], I. Pevzner[3], and Y. M. Kwon[1,2],
[1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]Cell and Molecular Biology Program, University of Arkansas, Fayetteville, AR, USA, [3]Cobb-Vantress Inc., Siloam Springs, AR, USA.

Bacterial chondronecrosis with osteomyelitis (BCO, also known as femoral head necrosis) is a common cause of lameness when observed in commercial broiler chickens worldwide. BCO represents substantial production loss and compromised welfare issues of chickens. The bacterial species or microbial communities underlying BCO pathogenesis remain to be fully characterized. This study was conducted to investigate blood microbiota associated with BCO leading to lameness. Blood samples were collected aseptically from the wing vein of normal (n = 240) and lame (n = 12) chickens followed by 16S RNA gene sequencing. MiSeq Illumina sequencing data were analyzed through closed reference observed taxonomic units (OTU) picking using Quantitative Insights into the Microbial Ecology (QIIME 1.9.1) pipeline. We found that the chicken blood microbiota were dominated by *Proteobacteria* (60.58% ± 0.65) followed by *Bacteroidetes* (13.99% ± 0.29), *Firmicutes* (11.45% ± 0.51), *Actinobacteria* (10.21% ± 0.37), and *Cyanobacteria* (1.96% ± 0.21), which together constituted 98.18% (SE ± 0.22) of the whole phyla. The bacterial communities consist of 30 to 40 OTU in the blood of broiler chickens, regardless of age and other environmental or host conditions. Linear discriminant (LEfSe) analysis revealed that *Staphylococcus*, *Granulicatella*, and *Microbacterium* were differentially abundant in BCO chickens compared with normal chickens. Additionally, phylogenetic investigation of communities by reconstruction of unobserved states (PICRUSt) followed by LEfSe analysis showed that genes related to DNA replication and repair or metabolism were significantly enriched in the blood microbiota of BCO chickens compared with normal chickens. The blood microbiomes analyzed in this study have significant implications on the health status of the broilers chickens, including BCO pathogenesis, as demonstrated in this study, as well as other disease conditions or stress conditions of broiler chickens.

112 Effects of virginiamycin and (or) a phytogenic feed additive on performance and intestinal health of broiler chickens.

H. M. Cervantes*[1,2], M. J. Da Costa[2], K. W. Bafundo[1], and G. M. Pesti[2],
[1]Phibro Animal Health, [2]University of Georgia.

A 42-d floor pen experiment was conducted at the University of Georgia Poultry Research Center with 1,920 Cobb-500 male broiler chickens to investigate the individual and combined effects of 20 ppm of dietary virginiamycin (VM) and a phytogenic feed additive (MP) at dietary concentrations of 150, 200, or 250 ppm and its potential interactions on performance and intestinal health. A 2 × 4 factorial experiment with 6 replicates of 40 birds each per treatment was carried out in a complete randomized block design with 2 dietary concentrations of VM (0 or 20 ppm) and 4 dietary concentrations of MP (0, 150, 200, and 250 ppm). Chickens and feed were weighed at 18, 32, and 42 d of age to calculate average body weight gain (BWG) and feed conversion ratio adjusted for mortality (AFCR). To investigate the effect of treatments on intestinal health, sections of the ileum from 12 birds from each treatment (2 birds per replicate) were collected on d 42. The intestinal sections were collected immediately after euthanasia and they were placed in 10% NBF and subsequently cut and stained with hematoxylin and eosin for histologic evaluation. Histomorphometric measurements evaluated included apical width, base width, villus height, crypt depth, and muscularis thickness. Analysis of variance was conducted on all data to determine if significant ($P < 0.05$) differences existed between treatments. On d 0, no differences were detected in BW; on d 18, 32, and 42, VM supplementation produced significantly better BWG ($P < 0.05$). Likewise, VM supplementation numerically improved AFCR on d 18 and 32, and the difference was significant ($P < 0.05$) at 42 d. There was a consistent trend toward higher BWG with all levels of supplementation of MP; however, the difference was only significant for the 200 ppm concentration at 32 d ($P < 0.05$). MP did not produce a significant improvement in AFCR at any period. The results from the histological examination showed that the broilers that consumed VM had significantly lower crypt depth ($P < 0.05$)—an indication of improved intestinal health—compared with control.

Session IV

113 Toward an understanding of fish gut tolerance to alternative feeds—The Trout-Grains Project.

J. Abernathy*[1], A. Brezas[2], T. Welker[1], K. Liu[3], F. Barrows[4], K. Overturf[1], and R. Hardy[2],
[1]USDA-ARS, Hagerman Fish Culture Experiment Station, 3059F National Fish Hatchery Rd, Hagerman, ID, USA, [2]University of Idaho, Hagerman Fish Culture Experiment Station, Hagerman, ID, USA, [3]USDA-ARS, Small Grains and Potato Germplasm Research Unit, Aberdeen, ID, USA, [4]USDA-ARS, Bozeman Fish Technology Center, Bozeman, MT, USA.

Rainbow trout (*Oncorhynchus mykiss*) and other salmonids are carnivorous. In aquaculture, manufactured diets for these species have conventionally relied heavily on fishmeal and fish oil to supply essential nutrients. As the global demand for farmed fish has increased substantially, along with the cost of marine-derived feed ingredients, fishmeal and fish oil-based feeds are neither economically nor environmentally sustainable. The use of alternative feed formulations in which fishmeal and fish oil are partially replaced with plant-based protein and oils has been an effective practice to a certain degree. However, in many farmed fish, total replacement of fishmeal with plant protein concentrates causes a cascade of pathological conditions, including intestinal enteritis, which leads to reduced growth and higher feed conversion ratios. After 15 years of selective breeding, the USDA-ARS in collaboration with the University of Idaho Aquaculture Research Institute has developed a strain of rainbow trout that thrives when fed high-soy, plant-protein-based diets. These fish do not develop distal intestinal enteritis, grow rapidly, and exhibit low feed conversion ratios. They also serve as a valuable research control, providing the ability to compare diet substitution in a healthy versus diseased model. We are interested in understanding the underlying pathophysiology and genetic changes associated with enteritis in an effort to improve the integration of sustainable plant-based diet formulations with genetic selection in rainbow trout farming as well as transferring this technology to other carnivorous fish species. Toward this goal, we are currently investigating the effects of diet formulation and genetic strains on such factors as gut metagenomics and meta-transcriptomics, fish tissue pathology, transcriptomics, and proteomics. The results of our efforts will be presented and discussed.

114 Use of a novel enzymatic strategy (Hemicell-L) to assess the effect on broiler intestinal health and productivity under commercial conditions in a German integration.

M. A. Martinez-Cummer*[1], G. González-García[2], and K. Poulsen[2],
[1]Elanco Animal Health, Greenfield, IN, USA, [2]S.A. Eli Lilly Benelux N.V., Antwerp, Belgium.

The use of phytase and fiber-digesting enzymes in European broiler diets is very common and their value is generally accepted. The main mechanism of these enzymes is to make nutrients more available to the birds. There is, however, a different group of enzymes with a very different mode of action, the energy-sparing enzymes, which spare energy by breaking down substrate in the feed that otherwise would provoke futile immune activation. Endo-1,4-β-D-mannanase is currently the only EU-approved enzyme of this kind. It degrades β-mannan fibers found in most vegetable feed ingredients (Hsiao et al., 2014). Consequently, it eliminates its anti-nutritional effect and prevents it from triggering an innate response that may adversely affect performance (Klasing et al., 1987). This so-called feed-induced immune response (FIIR) from β-mannans typically costs about 3% of performance (Daskiran et al., 2004; Anderson et al., 2006). The paper will present results of a 6-month field trial with Hemicell Liquid, conducted under common commercial production conditions by a German broiler integrator. The production observations from this field trial, together with its effects on intestinal integrity and animal welfare, will be presented.

115 Combining feed enzyme activities in poultry feeds: Adding energy-sparing enzymes to manage the effect of unnecessary feed-induced immune responses on broiler intestinal health and overall production performance.

M. A. Martínez-Cummer*,
Elanco Animal Health, Greenfield, IN, USA.

High feed and production costs have made the use of energy-releasing NSP enzymes common practice in poultry production. Producers are also under pressure to maximize health and efficiency in their flocks. Feed-induced immune response (FIIR), for instance, is one cause of unproductive energy expenditure, which occurs due to inappropriate action of the immune system. This occurs when the bird's immune system mistakes β-mannan fibers, which are found in feeds such as soybean, sunflower, and sesame meal, for harmful microorganisms and uses up vital energy mounting an immune response. Because soybean meal is a major protein source in feeds produced around the world, β-mannan is present in most feeds. It is estimated that 3% of a bird's metabolizable energy could be depleted by FIIR, which has significant consequences on overall productivity. The energy-sparing enzymes, such as β-mannanase with a very different mode of action, can be combined with all common energy-releasing feed enzymes to combat FIIR by breaking down the β-mannan fibers. The fibers are no longer recognized by the immune system and more energy is left available to the bird for growth, rather than being consumed by FIIR. Research has shown that significant improvements in health and production parameters seen with the inclusion of β-mannanase (Hemicell) in broiler diets is primarily due to the breakdown of β-mannan fibers to prevent unnecessary innate immune responses. This paper will discuss studies carried out to demonstrate the exact association between the β-mannan content of feed and the amount of immune system activation. Levels of immune system stimulation were measured with increasing levels of β-mannan-containing soybean meal. Studies to evaluate the effects of the β-mannanase enzyme on live performance, live weight uniformity, and health parameters at market age will also be discussed to show that FIIR is a reality and can affect broiler performance. The complementary benefits of adding energy-sparing enzymes with energy-releasing enzymes will be discussed.

116 Effects of grain processing on starch digestion and performance of feedlot cattle.

E. F. Schwandt*[1], J. J. Wagner[2], T. E. Engle[2], S. J. Bartle[1], D. U. Thomson[1], and C. D. Reinhardt[1],
[1]Kansas State University, Manhattan, KS, USA, [2]Colorado State University, Fort Collins, CO, USA.

Experiment 1 evaluated dry-rolled corn (DRC) particle size distribution and fecal starch content in finishing cattle in feedlots (n = 35) located in Midwestern US states. Samples of DRC, finishing diet, and freshly voided feces were collected from each feedlot. Average particle size of dry-rolled corn (n = 31) was 4,534 ± 899 µm with a range of 2,167 to 6,823 µm. Fecal starch content averaged 19.0 ± 6.5% with a range of 7.0 to 36.6%. Diet composition was evaluated for concentrations of grain by-product (27.8 ± 13.4%), roughage (8.9 ± 2.0%), and NDF (19.3 ± 4.3%). Experiment 2 utilized cross-bred yearling steers (n = 360; initial BW = 395 ± 33.1 kg) to evaluate the effects of DRC particle size in diets containing 20% (DM basis) wet distillers grains plus solubles on feedlot performance, carcass characteristics, and starch digestibility. Steers were allocated to 36 pens (9 pens/treatment; 10 animals/pen). Treatments were coarse DRC (4,882 µm), medium DRC (3,760 µm), fine DRC (2,359 µm), and steam-flaked corn (SFC, 0.35 kg/L). Final BW and ADG were not affected by treatment ($P > 0.05$). Dry matter intake was greater and gain:feed was lower ($P < 0.05$) for steers fed DRC versus SFC. There was a linear decrease ($P < 0.05$) in DMI in the final 5 wk on feed with decreasing DRC particle size. Fecal starch decreased (linear, $P < 0.01$) as DRC particle size decreased. No differences ($P > 0.10$) were observed among treatments for any of the carcass traits measured. In situ starch disappearance was lower for DRC versus SFC ($P < 0.05$) and increased linearly ($P < 0.05$) with decreasing particle size at 8 and 24 h. Reducing DRC particle size did not influence growth performance but increased starch digestion and influenced DMI of cattle on finishing diets. Reductions in DMI for more extensively processed grain may have been caused by prolonged subacute acidosis due to increasing rate of starch fermentation.

Key words: dry-rolled corn, fecal starch, particle size

Session V

117 Effect of organic acids and essential oils on enteric mucosa morphology and histology as indicators of gut health in poultry.

R. D. Malheiros*, V. M. B. Moraes, and P. R. Ferket,
Prestage Department of Poultry Science, NC State University, Raleigh, NC, USA.

There is growing interest in the use of alternatives to antibiotic growth-promoting (AGP) feed additives (organic acids, OA, or essential oils, EO) to manage gut health for poultry. Trials were done to evaluate these effects. In the first experiment, 200 day-old Nicholas toms were subjected to 2 levels of coccidia (challenged vs. unchallenged) and 2 levels of oregano essential oil (0 vs. 300 mg of Regano/kg of feed). Regano contains oregano oil in a soluble form. Each poult was gavaged at 3 d with either a 1-mL solution containing *Eimeria adenoeides* and *Eimeria meleagrimitis* (2,500 oocysts/bird) or 1 mL of water. Proximal ileum samples were taken for SEM and HIST analysis on 8 and 15 d. There were no significant treatment interaction effects on BW or rIW. *Eimeria* challenge reduced 14-d BW (366.4 vs. 410.6 g, $P < 0.0001$) and increased 15-d rIW (0.0836 vs. 0.0941, $P < 0.05$), but Regano had no significant effect on 14-d BW. Based on SEM evaluation, *Eimeria* challenge caused significant villi distress, whereas Regano enhanced mucin secretion and other morphological defense responses. In another experiment, EO from cashews and castor bean [Essential (Es), Oligo Basics USA, Cary, NC] was evaluated as an alternative to monensin (M) on early performance and gut morphology of poults. The 3 treatments consisted of a non-medicated control (C), dietary inclusion of 0.15% Es, and 66 ppm of M. Compared with C at 21 d, jejunum villi height and crypt depth was greater among Es-treated poults, whereas villi surface area was greater among M-treated poults. Es resulted in a positive growth performance and mucosal development of starting poults similar to that of M, indicating that it could be a potential non-pharmaceutical alternative to an ionophore such as monensin. A final experiment evaluated the effect of a protected OA and EO (300 g of Gallinat+/tonne, Jefo Inc., St. Hyacinthe, QC, Canada) on duodenum HIST of broilers in compared with 30 g of BMD/tonne. The protected OA-EO treatment resulted in significantly greater villi height and less morphological distress than the BMD treatment. The results of these experiments showed that OA and EO have a positive effect on intestinal mucosa morphology as indicators of gut health and demonstrate alternatives to antimicrobial pharmaceuticals.

118 Leaky gut and mycotoxins: Aflatoxin B_1 does not increase gut permeability in broiler chickens.

R. Galarza-Seeber*, J. D. Latorre, A. Wolfenden, B. Hargis, and G. Tellez,
University of Arkansas, Fayetteville, AR, USA.

We have previously shown that intestinal barrier function can be adversely affected by poorly digested diets or feed restriction, resulting in increased intestinal inflammation-associated permeability. Two experiments were conducted in broilers to evaluate the effect of 3 concentrations of aflatoxin B_1 (AFB1; 2, 1.5, or 1 ppm) on systemic fluorescein isothiocyanate-dextran (FITC-d; 3–5 kDa) levels, indicators of increased gut epithelial leakage and liver bacterial translocation (BT). In experiment 1, 240 day-of-hatch male broilers were allocated randomly in 2 groups, and each group had 6 replicates of 20 chickens (n = 120/group): Control feed or control feed + 2 ppm AFB1. In experiment 2, 240 day-of-hatch male broilers were allocated randomly in 3 groups, and each group had 5 replicates of 16 chickens (n = 80/group): Control feed; control feed + 1 ppm AFB1; or control feed + 1.5 ppm AFB1. In both experiments, chickens were fed starter or grower diet (d 7 to 21) ad libitum until the end of the trial at d 21. In both experiments, all chicks received an oral gavage of FITC-d (4.16 mg/kg) 2.5 h before collecting blood samples to evaluate passage of FITC-d. Liver sections were aseptically collected to determine BT on TSA plates. Cecal contents were collected to determine total counts (cfu/g) of gram-negative bacteria, lactic acid bacteria (LAB), or anaerobes by plating on selective media. Gut leakage of FITC-d was not affected by the 3 concentrations of AFB1 evaluated ($P > 0.05$). Interestingly, a significant reduction in BT was observed in chickens that received 2 and 1 ppm, whereas chickens that received 1.5 ppm showed only a numerical reduction in BT compared with control chickens. An increase ($P < 0.05$) in total aerobic bacteria, total gram-negative bacteria, and total LAB were observed in chickens fed 2 and 1.5 ppm of AFB1 compared with control and 1 ppm chickens. The integrity of epithelial gut barrier was not compromised after exposure to the mycotoxin.

119 Life cycle of probiotic *Bacillus* in the gastrointestinal tract.

M. Bernardeau*[1,2], M. J. Lehtinen[3], S. D. Forssten[3], and P. Nurminen[3],
[1]DuPont-Danisco, Industrial Biosciences, Animal Nutrition, Marlborough, United Kingdom, [2]QAIEA Research Unit, University of Caen, Caen, France, [3]DuPont Nutrition and Health, Kantvik, Finland.

Bacillus spp. are widely used in animal production for their probiotic properties such as digestibility improvement, health maintenance, immune modulation, and gut microflora modulation or growth improvement. Bacilli are fed to animals as spores due to their stability during feed processing and storage. However, microorganisms must be in a viable state to function as probiotics and compete for essential nutrients. The metabolic processes are essential for probiotic function in the host gastrointestinal tract (GIT) and for effective combination with other active substances such as enzymes. Considering that the probiotic screening process involves selection that is based on functional properties of the metabolically active strain (vegetative cell), and the sporulated form is convenient, easy to obtain and cost-effective, but does not constitute a beneficial property similar to metabolically active cells, it is of particular interest whether strains from *Bacillus* spp. are able to germinate in the GIT of animals. The fate of ingested *Bacillus* spores in the GIT of hosts (human and animals) need to be investigated, as well as the effective ratio of vegetative cells and spores that reach different parts of the GIT, the length of time that probiotic *Bacillus* persist in the gut after withdrawal from diet, and the influence of GIT physiology on germination potential. The *Bacillus* life cycle (especially probiotic *Bacillus*) has thus been reviewed in various intestinal environments, focusing on

germination and persistence data. There is clear evidence that *Bacillus* spores are able to germinate in GIT conditions. However, the germination process is highly dependent on the strain itself, GIT location, environmental conditions, maturity of the intestine, and conditions applied during the sporulation process before ingestion. Ingested spores of *Bacillus* will then transit through the GIT according to a ratio of vegetative cells to spores, which may vary in different gut sections according to transit time, gut motility, microbiota, environmental conditions (e.g., pH, oxygen level, nutrient availability), and the nature of the feed. Thus, it is suspected that a large amount of ingested spores may pass through the GIT and the metabolically inactive nature raises questions on their possible mode of action.

120 Effect of dietary supplementation of β-galacto-oligosaccharides on intestinal microarchitecture of broilers reared under cyclic heat stress.

S. Ashraf*, H. Zaneb, H. Rehman, S. Muti, and S. Ijaz,
University of Veterinary and Animal Sciences Lahore, Lahore, Punjab, Pakistan.

This study was designed to explore the potential role of β-galacto-oligosaccharides (GOS) in alleviation of detrimental effects of heat stress in broilers. Day-old broilers (n = 250) were randomly divided into 5 groups with 5 replicates containing 10 birds in each replicate. Two groups were non-supplemented: a thermoneutral (TN) and a heat-stressed (HS) group. Three groups were supplemented (GOS-0.1%, GOS-0.2%, GOS-0.5%). Birds in the HS and supplemented groups were exposed to cyclic heat stress from d 22 to 35 (35°C, 75% RH, 8 h/d). On d 35, birds (2/replica) were slaughtered to collect various segment of small intestine. The samples were processed for intestinal microarchitecture, intraepithelial lymphocyte (IEL) count, and goblet cell (GC) differentiation. Results showed that heat stress decreased ($P < 0.05$) villus height (VH), crypt depth (CD), and surface area (SA) in all intestinal segments compared with the TN group. The villi were taller ($P < 0.05$) in supplemented groups compared with the nonsupplemented HS group in duodenum, jejunum, and ileum. Similarly, crypts were deeper ($P < 0.05$) in the GOS-included groups compared with the heat-stressed group in jejunum and ileum. However, this effect could not be observed in the duodenum. Exposure of birds to cyclic heat stress significantly ($P < 0.05$) reduced the population of IEL compared with the TN group in all intestinal segments. Feeding GOS-0.1% elevated ($P < 0.05$) the IEL count compared with the other supplemented groups in the intestine. Heat stress reduced ($P < 0.05$) the count of the acidic GC in duodenum and jejunum but increased ($P < 0.05$) in the ileum compared with the TN group. Supplementation of GOS-0.2% increased the acidic GC count in the jejunum and ileum compared with the HS group. In conclusion, β-GOS can be a potential prebiotic candidate to supplement the poultry diet against some of the negative effects of heat stress.

Poster Presentations

P100 Daily feeding of *Lactobacillus animalis* improved performance and reduced mortality and morbidity associated with necrotic enteritis in broilers in two different model systems.

S. Lerner*[1], J. McNaughton[2], and G. Mathis[3],
[1]Nutrition Physiology Company LLC, Kansas City, MO, USA, [2]AHPharma, Salisbury, MD, USA, [3]Southern Poultry Research, Athens, GA, USA.

In system 1, 320 mixed-sexed broiler chicks were randomized into 20 floor pens. Birds in 10 pens received 1×10^8 cfu of *Lactobacillus animalis* (Poultrimax) in their feed daily. Birds in the other pens served as untreated controls. To built-up litter bedding, *Clostridium perfringens* (10^{7+}cfu/bird, considered to be a SEVERE stress level), *Eimeria acervulina* (10^4 cfu/bird), and *Eimeria maxima* (10^3 cfu/bird), were added on d 7 and 10 post-hatch. Weight gain, feed conversion, mortality, and intestinal lesion scores were compared between groups at d 21. Birds fed Poultrimax weighed more than controls (851.2 ± 5.6 g vs. 803.3 ± 5.9 g; $P < 0.05$), converted feed more efficiently (1.33 ± 0.01 vs 1.39 ± 0.01; $P < 0.05$), had lower mortality (3.12 ± 0.99% vs 9.38 ± 1.33%; $P < 0.05$), and had lower lesions scores (0.63 ± 0.05 vs 1.23 ± 0.05; $P < 0.05$). In system 2, 256 mixed-sexed broiler chicks were randomized into 32 battery cages. Eight cages were assigned to 1 of 4 treatments: non-medicated, unchallenged (NegCtrl; non-medicated, necrotic enteritis (NE)-challenged (PosCtrl); *L. animalis*-treated, NE-challenged (Poultrimax); and virginiamycin-treated, NE-challenged (Stafac 20, virginiamycin). On d 14, all birds were orally inoculated with ~5,000 oocysts of *E. maxima*/bird. All challenged birds were given a broth culture of *C. perfringens* 10^8 cfu/mL daily on d 19, 20, and 21. Birds were weighed by cage on d 0, 14, 21, and 28. Feed was weighed in on d 0 and remaining feed was weighed on d 14, 21, and 28, and the trial was terminated on d 28. On d 21, 3 birds from each cage were selected, killed, weighed, and examined for the degree of presence of necrotic enteritis lesions. For all variables except lesion scores, birds in the Poultrimax and Stafac groups performed better ($P < 0.05$) than birds in the PosCtrl group and poorer ($P < 0.05$) than birds in the NegCtrl group. In the post-challenge period (d 14 to 28), feed conversion, mortality, and lesion scores did not differ significantly between the Poultrimax and Stafac groups. In conclusion, daily feeding of a *L. animalis*-based probiotic improved performance and ameliorated the mortality and morbidity associated with necrotic enteritis in growing broilers.

P101 Effect of a *Bacillus* direct-fed microbial candidate on nutrient digestibility, gut permeability, bone quality, and growth performance in broilers consuming a sorghum-based diet.

J. D. Latorre*[1], R. E. Wolfenden[2], J. L. Vicente[2], A. D. Wolfenden[1], R. Galarza-Seeber[1], V. A. Kuttappan[1], L. R. Bielke[1], B. M. Hargis[1], and G. Tellez[1],
[1]University of Arkansas, Department of Poultry Science, Fayetteville, Arkansas, USA, [2]Pacific Vet Group, Fayetteville, Arkansas, USA.

Alternative grains such as sorghum are used worldwide in poultry diets. However, sorghum contains antinutritional factors such as phytate, polyphenols and kafirins, that affect nutrient digestibility and performance. The objective of this study was to evaluate the effect of a *Bacillus* direct-fed microbial (DFM) on apparent nutrient digestibility, gut permeability, bone quality, and growth performance. In the present study, 2 independent experiments were conducted using day-of-hatch chicks randomly assigned to 2 different treatment groups: Control (without DFM) or *Bacillus*-DFM (10^6 spores/g of feed). In experiment 1, 4 replicates of 12 broilers were used per group. Body weight was evaluated weekly. At d 21, samples of ileal content, blood, and liver tissue were obtained from 6 broilers per replicate to evaluate apparent nutrient digestibility (TiO_2) and gut permeability (serum FITC-d and bacterial translocation). In experiment 2, 8 replicates of 20 broilers were used per group. Body weight, feed intake, and feed conversion ratio were determined weekly. At d 28, both tibias were evaluated for bone strength (kg/mm^2) and bone composition (% ash, Ca, P) from 2 broilers per replicate. In experiments 1 and 2, significant improvements were observed in performance parameters in chickens consuming the DFM compared with the control group. In experiment 1, the DFM-supplemented diet improved apparent nutrient digestibility when compared with the non-supplemented group ($P < 0.05$). Additionally, there was a reduction in all gut permeability measurements in the *Bacillus*-DFM group compared with control ($P < 0.05$), indicating reduced gut leakage and suggesting improved gut health. In experiment 2, bone quality was improved ($P < 0.05$) in the DFM group compared with the control group. These findings suggest that inclusion of this selected *Bacillus*-DFM candidate in sorghum-based diets enhanced nutrient digestibility, reduced gut permeability, and improved bone quality while enhancing growth performance in broiler chickens.

P102 Evaluation of a *Bacillus* spp. direct-fed microbial candidate for aflatoxin B_1 biodegradation in broiler chickens.

R. Galarza-Seeber*, J. D. Latorre, A. Wolfenden, B. Hargis, and G. Tellez,
University of Arkansas, Fayetteville, AR, USA.

Aflatoxins are commonly found in cereals worldwide and bring significant threats to the food industry and animal production. The limitations of present physical and chemical methods to decrease aflatoxin in feed ingredients have encouraged research on biological methods of degradation. The ability of this direct-fed microbial (DFM) candidate to biodegrade aflatoxin B_1 (AFB1) in situ was evaluated in broilers by measuring body weight (BW), body weight gain (BWG), feed intake (FI), and feed conversion ratio (FCR) in 2 independent experiments. In experiment 1, 480 day-of-hatch male broilers were allocated randomly to 4 groups, each with 6 replicates of 20 chickens (n = 120/group): control feed; control feed supplemented with DFM (10^6 spores/g); control feed containing 2 ppm AFB1; control feed containing 2 ppm AFB1 and supplemented with DFM. In experiment 2, 480 day-of-hatch male broilers were allocated randomly to 6 groups, each with 5 replicates of 16 chickens (n = 80/group): control feed; control feed supplemented with DFM (10^6 spores/g); control

feed containing 1 ppm AFB1; control feed containing 1 ppm AFB1 and supplemented with the DFM; control feed containing 1.5 ppm AFB1; control feed containing 1.5 ppm AFB1 and supplemented with the DFM. In both experiments, chickens were fed starter and grower (d 7–21) diets ad libitum. Performance was recorded weekly. In experiment 1, both groups containing 2 ppm AFB1 had reduced BW and BWG at 14 and 21 d of age ($P < 0.05$). Similarly, AFB1 decreased BW and BWG in experiment 2 in a concentration-dependent manner. The DFM used in these experiments did not improve performance of broilers fed any concentration of AFB1. However, DFM treatment improved BW and BWG in feed not containing AFB1 compared with controls in experiment 1, although these results were not observed in experiment 2. Studies to evaluate more realistic concentrations of AFB1 in the field (500 and 50 ppb) are currently being evaluated.

P103 Involvement of microRNAs in calf intestinal growth and development and probable modulatory role of *Saccharomyces cerevisiae*.

E. M. Ibeagha-Awemu*, P.-L. Dudemaine, J. Chiquette, G. Talbot, and N. Bissonnette,
Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, Quebec, Canada.

Feed additives including probiotics are administered to stimulate immune maturation in the early period of calf growth. In this study, we examined *Saccharomyces cerevisiae boulardii* (SCB) survival and possible modulation of microRNA (miRNA) expression in the gastro-intestinal tract (GIT) of calves. MicroRNAs are short molecules of ~22 nt with regulatory roles in many biological processes. Twenty-four 2- to 7-d-old calves were allocated equally into control treatment [CTL; milk replacer + starter diet (from wk 3)] and SCB *CNCM I-1079* treatment (CTL diet + 7.5×10^9 cfu/L milk replacer + 3×10^9 cfu/kg of feed). Feces were sampled weekly for testing SCB viability. Four calves per treatment were euthanized on d 33 (preweaning) and d 89 (postweaning). Total RNA from rumen and ileum tissues was subjected to miRNA sequencing. Fecal SCB counts decreased linearly ($P = 0.005$) from 1.7×10^6 cfu/g on d 1 to 2.3×10^5 cfu/g on d 57 (1 wk after weaning) and increased to 6.0×10^5 cfu/g on d 89 ($P = 0.01$). Counts of SCB were greater in the feces (3.8×10^5 cfu/g) than in the ileum content (4.8×10^4 cfu/g; $P = 0.05$) before weaning (d 33), whereas there was a tendency ($P = 0.06$) for a greater SCB count in colon (4.8×10^5 cfu/g) than in the rumen (9.7×10^4 cfu/g) after weaning (d 89). In total, 388 (including 81 novel) and 295 (including 35 novel) miRNAs were identified in ileum and rumen, respectively; 251 known and 33 novel miRNAs were shared between tissues; and 40 and 153 miRNAs were differentially expressed (FDR < 0.05) between d 33 and d 89 in ileum and rumen, respectively. The SCB treatment affected (FDR < 0.05) the expression of one miRNA each on d 33 and d 89 in rumen and 2 miRNAs on d 33 in ileum. Functional enrichment of differentially expressed miRNAs showed roles in cellular growth and development (cellular growth and proliferation, cell death/survival, cellular movement, cell cycle, and cell morphology). Furthermore, differentially expressed miRNAs are involved in pathways of inflammatory response and cell-to-cell signaling. Results suggest that miRNAs may play roles in the growth and development of the gastrointestinal tract of calves. Our data further suggests that SCB can survive and grow in the gastrointestinal tract of calves with potential effects on miRNA regulatory activities.

P104 Sugar cane bagasse affects the fungal community dynamic in the sheep rumen.

E. M. Romagnoli*[1], C. Dunlap[2], A. L. Abdalla[3], and R. Mendes[1],
[1]Embrapa Environment, Laboratory of Environmental Microbiolog, Jaguariuna, Sao Paulo, Brazil, [2]USDA-Agricultural Research Service, National Center for Agricultural Utilization Research, Peoria, IL, USA, [3]University of Sao Paulo, Center for Nuclear Energy in Agriculture, Piracicaba SP, Brazil.

Sheep have evolved a symbiotic host-microbe relationship with a complex microbial community inhabiting their rumen which facilitates the conversion of lignocellulose biomass into their main energy source. Considering that diet is one of the main drivers shaping the structure of the rumen microbiome, we investigated the effect of sugarcane bagasse in the rumen fungal community dynamic using the nuclear ribosomal internal transcribed spacer ITS region (ITS2) amplicon sequencing. We assessed 3 rumen-cannulated adult male sheep (*Ovis aries*) fed a diet composed of 30% concentrate and 70% roughage (control treatment) and another 3 sheep fed the same diet but with 14% of the roughage portion replaced by sugarcane bagasse. Partially digested fiber was collected sampled at 3 h and 15, 30, 45, and 60 d after starting the experiment. Total genomic DNA was extracted from 30 independent samples (2 treatments × 3 replicates × 5 times points) for downstream analysis. Overall, the 2 dominant phyla were Chytridiomycota (91%) and Ascomycota (4%). The most abundant fungal genus was Piromyces (41%) followed by Neocallimastix (18%), Orpinomyces (14%), and Anaeromyces (11%). Principal coordinate analysis showed that the fungal community was significantly different between treatments (control × bagasse) at 60 d. Neocallimastix, Orpinomyces, Anaeromyces, and Piromyces were the top dynamic fungal genera that significantly increased in relative abundance in the treatment with sugarcane bagasse after 60 d. These results indicate that a minor replacement in the diet roughage portion influences the dynamic of specific fungal taxa. Study was supported by FAPESP 2012/03848-8, 2012/24588-4 and 2014/00448-4.

P105 A comparison of fungal and bacterial populations in broilers from high- and low-producing farms.

J. Byrd*,
USDA, ARS, Food and Feed Safety Research Unit, College Station, TX, USA.

Fungus and yeast are consistent members of animal microflora that are poorly understood as related to the production of poultry. Fungi, like bacteria in the past, have been associated with the onset of disease. Little attention has been given to the beneficial effects of fungi (and particularly various yeasts) with regard to food safety and especially with the gastrointestinal tracts of food-producing animals. In this study, we surveyed the changes that occur in both fungal and microbial populations in health commercial broilers. Four complexes were selected from the southern United States over a 12-mo period. On each complex, 5 commercial houses were selected from high-producing farms and

5 houses from low-producing farms for a total of 10 houses per complex. For microbiome analysis, crop, duodenum, jejunum and cecal, samples were collected from each bird (n = 5/house) and 10 farms per complex on d 30 to 36. These samples were used for microbiome analysis by deep sequencing of the V1-V3 regions with MiSeq. Analysis of similarity (ANOSIM) of the fungal samples showed that the groupings based on age and sample type were statistically significant ($P \leq 0.05$). In the cecal samples collected from the high- and low-producing farms, no differences were observed between the cecal bacterial populations. However, dramatic changes were observed in the fungal populations of Cryptococcus, Candida, Malassezia, and Ascomycota genera in the ceca of birds sampled on the high- versus low-producing farms. Understanding the fungal and bacterial changes that occur in gastrointestinal tracts of commercial poultry may help us develop a gut health model to target areas for potential production and poultry health improvements and that may be utilized in the development of intervention strategies to control foodborne pathogens.

P106 Genetic relatedness between avian pathogenic *Escherichia coli* recovered from broiler breeders and chicks reveals potential for vertical transmission.

J. Lambrecht*[1], F. Delago[1], K. Gibbs[2], T. Horne[3], and E. Galbraith[1],
[1]DuPont Nutrition and Health, Waukesha, WI, USA, [2]DuPont Industrial Biosciences, Marlborough, United Kingdom, [3]Chemuniqué, Randburg, South Africa.

Colibacillosis, caused by avian pathogenic *Escherichia coli* (APEC), contributes to major economic losses throughout the poultry industry worldwide. APEC can be found in birds of all ages, including newly hatched chicks. A potential route of infection in birds this young is vertical transmission. The aim of this study was to investigate the similarity of genotypes and virulotypes in APEC isolates obtained from broiler breeders and their offspring. *Escherichia coli* was recovered from the gastrointestinal tract of 27 breeders (135 isolates) and 90 day-old chicks (DOC; 71 isolates) originating from 3 houses at producer 1 farms A and B, and 5 houses at producer 2 farms C and D. Multiplex toxin gene PCR was used to screen isolates for 5 virulence-associated genes (VG): *iss*, *iucC*, *tsh*, *cvaC*, and *irp2*. Isolates containing 2 or more of the 5 genes were considered APEC. Breeder APEC isolates most commonly contained 3 VG with *irp2* occurring most frequently, whereas the majority of DOC isolates had 2 VG. with *cvaC* occurring most often. All isolates from farm A house 2 DOC had the virulotype *cvaC*/*irp2*. This pattern was also identified in several of the breeders from that same farm; however, the most prevalent virulotype in producer 1 breeders was *iucC*/*tsh*/*cvaC*/*irp2*. The virulotype *tsh*/*cvaC*/*irp2* was found in all APEC isolates from DOC and 70% of those from breeders at farm D house 1, and was the most common virulotype across all breeders from producer 2. Random amplified polymorphic DNA (RAPD)-PCR was used to determine genetic similarity among APEC isolates. All DOC APEC isolates grouped most closely with isolates from the same producer; additionally, the majority exhibited greater than 80% similarity to breeder isolates from the same farm. Two clusters of farm D house 1 isolates showed more than 90% similarity among DOC and breeder APEC isolates that also contained the same VG profile. These genetic similarities between breeder and DOC isolates from the same farm illustrate the potential transmission of APEC from broiler breeders to their progeny.

Key words: broiler breeder, day-old chick, APEC

P107 Investigation of avian pathogenic *Escherichia coli* (APEC) virulence-associated genes from US poultry isolates.

F. Delago*, C. Urbain, J. Lambrecht, S. Gebert, and E. Galbraith,
DuPont, Waukesha, WI, USA.

Avian pathogenic *Escherichia coli* (APEC) virulence definition depends on the presence of 2 or more virulence genes (VG), including but not limited to *iuc*C, *tsh*, *iss*, *cvaC*, and *irp*2. These genes are not universally prevalent among APEC, which results in potentially diverse pathogenicity mechanisms. The objective of this study was to conduct a thorough investigation of 9,601 broiler, layer, and turkey *E. coli* isolates from the eastern half of the United States for their virulence-associated genes and patterns. The *iss* gene was the least prevalent at 29% of all isolates, whereas *iucC*, *tsh*, *cvaC*, and *irp*2 were equally present at around 40%, regardless of poultry species. Investigation of unique gene patterns of APEC isolates revealed that isolates with only one gene were most prevalent in broilers and layers and isolates containing all 5 genes were least prevalent in all 3 poultry species. Isolates containing 2 genes were present in 36.0, 43.5, and 29.5% of broilers, layers, and turkeys, respectively. The most prevalent patterns of 2 genes were *iucC*/*irp*2 in broilers, *iss*/*cvaC* and *iucC*/*tsh* in layers, and *tsh*/*cvaC* in turkeys. Prevalence of APEC isolates containing 3 genes were detected at 31.4, 27.2, and 33.1% in broilers, layers, and turkeys, respectively. The most prevalent patterns of 3 genes were *iss*/*tsh*/*cvaC* in broilers and turkeys, *iss*/*iucC*/*cvaC* in layers, and *iucC*/*tsh*/*cvaC* in turkeys. Prevalence of APEC isolates containing 4 genes were detected at 25.1, 29.3, and 30.1% in broilers, layers, and turkeys, respectively. The most prevalent pattern of 4 genes was *iss*/*iucC*/*tsh*/*cvaC* in both broilers and turkeys and *iss*/*tsh*/*cvaC*/*irp*2 in layers. The unique combination of all 5 genes, *iss*/*iucC*/*tsh*/*cvaC*/*irp*2, was only detected at 7.5 and 7.3% in broilers and turkeys, respectively, whereas this pattern was not observed in layers. Overall, the pattern of virulence genes was not consistent in broilers or layers; however, in turkeys, the *tsh* and *cvaC* genes were consistently present in the highest prevalent patterns of APEC isolates.

P108 Effect of organic selenium on the chicken cecal microbiome equilibrium and *Campylobacter jejuni* carriage.

A. Thibodeau, P. Fravalo*, and A. Letellier,
Université de Montréal, Saint-Hyacinthe, Québec, Canada.

In-feed nonantibiotic supplements are increasingly being used and developed for animal food production to limit the use of antibiotics in animal husbandry. Selenium has been successfully used in chicken to increase the animal performances, although the effect of this additive on the chicken gut microbiome is unknown. Its effect on foodborne pathogens is also not studied. It is especially important in the case of the foodborne pathogen *Campylobacter jejuni*, as it has been shown that selenium is important for this bacterium to achieve an efficient colonization

of its chicken host. We therefore assessed the effect of organic selenium (yeast) on the chicken cecal microbiome and *C. jejuni* carriage. In this experiment, 2 chickens groups, inoculated or not with *C. jejuni* at 15 d of age, were fed a basal mashed diet with the following in-feed additives: (1) no additive; (2) selenium-yeast (0.3 ppm). Birds were raised in a level 2 animal facility with the *C. jejuni* positive and negative birds housed in different rooms. This experiment was replicated once. Birds were weighed weekly. *C. jejuni* colonization was followed using plating on mCCDA. Serum glutathione peroxidase (GPX) levels were also monitored. At 35 d of age, 8 birds per groups were ethically euthanized. Birds fed selenium were heavier in 1 of the 2 experiments. *C. jejuni* levels were decreased in one experiment, for the groups receiving selenium. Serum GPX levels were lowered in the groups that received selenium and this in both experiments. The microbiome of all birds is currently being analyzed by MiSeq 16S rDNA sequencing to asses any effect of the additive on the chicken microbiome. So far, in high sanitary conditions (level 2 facility) and in healthy birds, we conclude that bird performance and *C. jejuni* carriage were slightly and inconsistently affected when selenium-yeast was used.

P109 Metagenomic analysis of the bovine hindgut from *Salmonella* Kentucky and Cerro-shedding dairy cows.

B. Haley*, J. Karns, and J. Van Kessel,
Environmental Microbial and Food Safety Laboratory, Beltsville Agricultural Research Center, Agricultural Research Service, US Department of Agriculture, Beltsville, MD, USA.

In the United States, *Salmonella enterica* ssp. *enterica* serovars Kentucky and Cerro are frequently isolated from dairy cows that appear asymptomatic. Although they are not major contributors to the salmonellosis burden, these serovars have been implicated in human clinical cases in recent years. To investigate the diversity of the bovine hindgut as well as associations between *Salmonella* status and bacterial diversity, the 16S rRNA metagenomes of fecal samples from cows on a single dairy farm were sequenced. Fecal grab samples were collected from 20 dairy cows on 5 different sampling dates. The *Salmonella* status of the cows was determined by enrichment of feces in tetrathionate broth followed by real-time PCR to detect the *invA* gene. For metagenomic analysis, genomic DNA was extracted from each fecal sample followed by 2 rounds of conventional PCR with 16S rRNA specific primers and Nextera DNA indices, respectively. Paired-end sequencing of 16S rRNA amplicons was performed on the Illumina MiSeq platform using 500 cycle V2 cartridges. Sequencing reads were analyzed with QIIME version 1.8.0. A high level of variability was observed between samples, demonstrating that the microbial profiles of bovine hindguts within the same herd are diverse. To determine whether *Salmonella* status, sampling year, or sampling group explained a significant amount of the variation in microbial diversity, we performed a constrained ordination analyses (distance-based RDA; rb-RDA) on the unifrac distance matrix produced with QIIME. Results indicated that there was a significant difference in the microbial diversity associated with sampling year and date but not for *Salmonella* status.

P110 Effect of stress events on mucosal permeability, bacterial translocation, and lameness in broilers.

L. R. Bielke*[1], J. C. Bielke[1], V. A. Kuttappan[2], E. A. Vicuña[2], A. Al-Ogaili[2], J. D. Latorre[2], B. D. Mahaffey[2], B. M. Hargis[2], and G. I. Tellez[2],
[1]Ohio State University, Department of Animal Sciences OARDC, Columbus, OH, USA, [2]University of Arkansas, Poultry Science Department Poultry Health Laboratory, Fayetteville, AR, USA.

Previous reports indicate that enteric inflammation and associated permeability may play a crucial role in pathogenesis of multiple diseases associated with lameness in poultry, including bacterial chondronecrosis with osteomyelitis, turkey osteomyelitis complex, and spondylolisthesis. Additionally, dexamethasone has been shown to increase mucosal permeability, measured by increased fluorescein isothiocyanate-dextran (FITC-d) in serum and bacterial levels in liver. In experiment 1, treatments consisted of negative control, 8-h water withdrawal immediately before sampling, dexamethasone in feed (d 4–11), rye-based diet (d 7–11), and 15% dried distillers grain with solubles (DDGS). Liver bacteria and FITC-d were measured on d 11. Dexamethasone, rye-based diet, and water withdrawal resulted in the highest levels of serum FITC-d, and percentage incidence of positive bacterial translocation to liver were 80, 73.3, and 60%, respectively, compared with only 26.7% for DDGS group and 33.3% for control. In experiment 2, all chicks were exposed to a mild cold stress (30°C for 6 h) at 3 d of age plus an inflammation-inducing treatment, followed by *Enterococcus cecorum* (EC) challenge on d 11, except for controls, which received only the cold stress treatment. On d 11, serum FITC-d levels were measured, and EC was recovered from the free thoracic vertebrae region on d 15. Birds were monitored for lameness through d 70. Serum FITC-d and EC recovery were highest ($P < 0.05$) in dexamethasone-treated birds and increased on the rye-based diet and DDGS diet . However, by d 70, there were no differences in total lameness or occurrence of spondylolisthesis, and incidence of ascites compared with control groups. This suggests that although dexamethasone and dietary treatments increase mucosal permeability, low level and perhaps undetectable stressors such as temperature change and short-term water withdrawal may be sufficient for inducing meaningful amounts of decreased enteric integrity. These experiments stress the importance of early management on development and maintenance of enteric integrity and long-term health of flocks.

P111 The effect of sericea lespedeza on coccidia-infected chickens.

L. Trinh*,
Louisiana State University, Baton Rouge, LA, USA.

Parasites cause considerable economic losses in all fields of agriculture. In the poultry industry, coccidia infections are of major significance, costing billions of dollars every year. Coccidiosis is a disease that is caused by protozoan parasites of the genus *Eimeria*, developing within the intestine of most domestic and wild animals and birds. Drug resistance calls for new methods of alternative control. Natural alternatives would be faster to produce and would allow organic farmers access to parasitic control without risk to the organic label. Sericea lespedeza is a legume that was introduced to the United States in the 1900s for

erosion control. It is found throughout the southern coast and contains condensed tannins. Research shows that condensed tannins are effective against various parasites such as nematodes and coccidia in small ruminants. This study will test the effects of sericea lespedeza (SL) on chickens infected with a live attenuated vaccine containing coccidia. Three hundred Ross 708 broilers will be divided into 6 groups (negative control, positive control, and 0, 5, 10, and 15% SL). Chickens will be fed for 18 d in battery cages. All groups, except the negative control, will be infected with a high dose of Coccivac-D2 at d 4 via gavage (0.5 mL of water-diluted vaccine, approximately 100,000 oocysts per bird). Positive control will compare a coccidostat (BioCox) with SL. Sericea will be fed to the chickens from d 0 to 18. Data analysis will comprise fecal oocyst counts, pen weights, feed conversion, and lesions. Significance in data will be calculated using a one-way ANOVA with Fisher's least significant difference (LSD) test; significance will be set at $P < 0.05$. The study is currently in progress and data will be presented at the symposium.

P112 Effect of seasonality in the intestinal integrity of broilers in Northern European countries in the period 2012–2014: Results of the Elanco Health Tracking System.

A. Zocche* and G. Garcia,
Elanco Animal Health, Greenfield, IN, USA.

For a long time, the poultry industry has been discussing if seasonality might influence the intestinal integrity in broiler flocks and if such an effect on intestinal integrity could be effectively measured. The countries chosen for this retrospective study were the United Kingdom, Northern Ireland, Germany, the Netherlands, and Poland. The criteria used were climate and weather. Summer and winter months in a 3-yr period between January 2012 and December 2014 were compared. The Intestinal Integrity Index (I^2 Index) is an evaluation of the intestinal tract for evidence of intestinal damage that can result in performance loss. It is a cumulative assessment of all enteric categories evaluated by a methodology called Health Tracking System (HTS). The difference in the I^2 index between winter and summer groups resulted in an improvement for the winter period of +0.44 points ($P < 0.01$). A difference of +22.04 g between the winter and the summer season was observed when the average live body weight (Avg. LBW) of both groups was compared ($P < 0.01$). No significant difference was observed in the age of the groups at the time of the postmortem analysis. Seasonality was estimated to have a remarkable effect of €20/1,000 kg of LBW for the 3-yr period 2012- 2014, or the equivalent of €16,000/yr for every million broilers of 2.4 kg Avg. LBW. Losing 1 point in I^2 Index in the Northern European region between 2012 and 2014 was estimated to be approximately −4.5 €cents/kg of LBW at the producer level. A multifactorial approach for a better intestinal integrity protection should be implemented during the summer. Further investigations need to be performed to better understand this effect and to identify which gastrointestinal changes vary between seasons.

Table 1 (Abstract P112).

Season	Monthly interval	No. of birds examined with HTS
Winter	Jan-May and Oct-Dec (8 mo)	16.275
Summer	Jun-Sep (4 mo)	8.918

			Price paid, adjusted to avg. LBW (€/kg)	
	I^2 Index (points)	Avg. LBW (kg)	Minimum	Maximum
Summer	93.62	1.160	1.08	1.10
Winter	94.06	1.182	1.10	1.12
Difference	−0.44	−0.022	−0.02	−0.02

P113 Systems kinomics reveal that *Salmonella enterica* Enteritidis modulates host immune signaling pathways in the cecum of chickens that are associated with the establishment of persistent infections.

M. H. Kogut*[1], C. L. Swaggerty[1], R. Salvaraj[2], and R. J. Arsenault[3],
[1]USDA-ARS, College Station, TX, USA, [2]Ohio State University, Wooster, OH, USA, [3]University of Delaware, Newark, DE, USA.

Nontyphoidal *Salmonella enterica* induce an early pro-inflammatory response in chickens. However, the response is short-lived, asymptomatic of clinical disease, resulting in a persistent colonization of the gastrointestinal tract that transmits infections to naïve hosts via fecal shedding of bacteria. The underlying mechanisms that allow the bacteria to persistently colonize the ceca of chickens are unknown. We hypothesize that alterations in the host immune signaling pathways mediate the induction of an anti-inflammatory or tolerogenic response. Using chicken-specific kinomic immune peptide arrays and quantitative RT-PCR of infected cecal tissue, we profiled the host immune response in chickens infected with *Salmonella enterica* serovar Enteritidis in a persistent infection model (4–14 d post-infection). We have outlined the induction of a tolerogenic response in the cecum of chickens infected with *Salmonella* Enteritidis beginning around 4 d after primary infection. The tolerogenic response is induced by a series of phosphorylation-mediated changes in immune signaling pathways in the ceca of chickens during the development of a persistent *Salmonella* infection. The tolerance is characterized by alterations in T-cell signaling

[dephosphorylation of phospholipase c-γ1, dephosphorylation of NF-κB, phosphorylation of nuclear factor of activated T cells (NFAT), dephosphorylation of multiple MAPK and mTOR signaling pathways (increased phosphorylation of AMPK) and blockage of IFN-γ protection through the disruption of the JAK-STAT signaling pathway (dephosphorylation of JAK2, JAK3, and STAT4)]. Further, the tolerogenic response was characterized by a dramatic reduction in IFN-γ mRNA expression through the 4- to 14-d post-infection period of these experiments. These studies describe specific immune phenotypic changes in the avian cecum of *Salmonella* Enteritidis-infected chickens that influence the host responsiveness, resulting in the establishment of persistent colonization. The altered tissue protein kinases also represent potential targets for future antimicrobial compounds that may decrease *Salmonella* loads from the intestines of food animals before going to market.

P114 Comparison of immunometabolic response in turkeys orally inoculated with *Salmonella* Heidelberg.

R. J. Arsenault*[1], M. H. Kogut[2], H. He[2], and K. Genovese[2],
[1]University of Delaware, Department of Animal and Food Sciences, Newark, DE, USA, [2]United States Department of Agriculture, College Station, TX, USA.

The *Salmonella* Heidelberg serovar is of increasing concern especially given the recent cases of contaminated poultry flocks and resultant zoonotic infections in humans. Both chickens and turkeys are susceptible to *Salmonella* Heidelberg, resulting in disease and contaminated food products. Individual turkeys can display a differential susceptibility to *Salmonella* Heidelberg colonization, and we hypothesize this may be due to differential responses to the bacteria. To study this, we used the turkey-specific immunometabolism kinome peptide microarray recently developed and reported by our laboratory. Turkeys hatched from 3 different flocks were inoculated with *Salmonella* Heidelberg, 7 d post-hatch. Five days post-infection, the turkeys were killed, and cecal contents, cecal tissue, and liver tissue was collected. Besides non-inoculated controls, cecum and liver from 2 birds from each flock that were *Salmonella* Heidelberg positive, liver from 1 bird from each flock that was *Salmonella* Heidelberg negative, and cecum from 1 bird from one flock that was *Salmonella* Heidelberg negative were assayed. We observed significant differences in the kinome profiles of inoculated turkeys found to be positive for *Salmonella* Heidelberg and those shown to be negative. In the inoculated but *Salmonella*-negative cecum, we observed changes in the phosphorylation state of metabolism intermediates, specifically those involved in acetyl-coA and fatty acid metabolism. When comparing the samples of liver where the *Salmonella* was able to leave the gut and invade the organ and the liver that was free of *Salmonella*, we observed similar levels of differential phosphorylation of immune response pathways relative to control, but distinct phosphorylation of key carbohydrate and fatty acid metabolic pathways. This study represents the first use of a kinome approach to study the host-response to a pathogen in turkeys. This is also the first reported use of the novel turkey-specific kinome immunometabolism peptide array technology. By comparing birds that were colonized with those that were not, we can find the key differences that can lead to a resistant turkey or an intervention that can aid in the clearance of *Salmonella*.

P115 Effects of supplementation of probiotics (*Bacillus*, *Lactobacillus*, *Aspergillus niger*) and prebiotics (chicory, rice bran) on growth performance, nutrient digestibility, meat quality, excreta microbiota, hematological profile, and excreta noxious gas emissions in broilers.

W. Yun*, H. S. Choi, H. G. Choi, Y. I. Choi, and J. H. Cho,
Chungbuk National University, Cheongju, Chungbuk, South Korea.

We introduce a feed supplement with probiotics (*Bacillus* at 1×10^9 cfu; *Lactobacillus* at 1×10^8 cfu; *Aspergillus niger* at 1×10^7 cfu) and prebiotics (chicory and rice bran as dietary fiber). This experiment was conducted to investigate the effects of supplementation of probiotics and prebiotics on growth performance, nutrient digestibility, meat quality, excreta microbiota, hematological profile, and excreta noxious gas emissions in broilers. A total of 714 one-day-old mixed-sex Ross 308 broiler with initial BW of 40 ± 0.69 g were used in a 5-wk trial. Birds were randomly allotted to 7 treatments with 6 replicates per treatment and 17 broilers per pen. Dietary treatments (T) were (1) control; (2) T1 + 0.2% probiotics; (3) T1+ 1% chicory; (4) T1 + 1% rice bran; (5) T1 + 0.2% probiotics + 1% chicory; (6) T1 + 0.2% probiotics + 1% rice bran; and (7) T1 + 0.2% probiotics + 1% chicory + 1% rice bran. During d 1 to 14, broilers fed the T5, T6, and T7 diets had greater body weight gain (BWG) compared with those fed the T1 diet ($P < 0.05$). Moreover, probiotics included treatments had higher BWG compared with the probiotics-free diets ($P < 0.05$). Broilers fed the T7 diet increased average daily gain (ADG) compared with the T1 diet, whereas feed conversion ratio (FCR) was decreased in the T7 diet compared with the T1 diet ($P < 0.05$). Dry matter (DM) digestibility was increased in the T5, T6, and T7 diets compared with the T1 diet ($P < 0.05$). Boilers fed probiotics-based diets improved dry matter (DM) digestibility compared with probiotics-free diets ($P < 0.05$). Excreta *Lactobacillus* counts were increased in the T7 diet compared with the T1 diet ($P < 0.05$). Broilers fed probiotics diets had higher excreta *Lactobacillus* counts compared with the probiotics-free diets ($P < 0.05$). In conclusion, supplementation with 0.2% probiotics and 1% dietary fiber as prebiotics could improve the growth performance, DM digestibility, and excreta *Lactobacillus* counts in broilers.

P116 Effects of direct-fed microbials on growth performance, gut microbiota, and pathogen resistance in *Litopenaeus vannamei* shrimp.

J. Barnes*[1], J. P. Gorsuch[1], M. S. Showell[1], C. L. Kitts[2], A. L. Lawrence[3], and R. S. Carpenter[1],
[1]BiOWiSH Technologies Inc., Cincinnati, OH, USA, [2]California Polytechnic State University, San Luis Obispo, CA, USA, [3]Ralco Animal Nutrition, Marshall, MN, USA.

Direct-fed microbials (DFM) by definition are products purported to contain live (viable) microorganisms. The effects of a DFM containing bacteria from the genera *Bacillus* and *Lactobacillus* on shrimp growth, survival, and digestive tract microbiome was studied. Upon termination of the growth study at 53 d, the following increases were observed: (1) 48.5% increase in shrimp size, (2) 83.0% increase in total biomass, and (3) 60% increase in survivability. These differences were not significant. Frozen shrimp from each treatment at termination were used for bacterial

taxonomic profiling. Intestines of the shrimp were extracted and pooled for each treatment. DNA was extracted from the samples, and taxonomic profiles of bacteria were created using a terminal restriction fragment length polymorphism (TRFLP) analysis. The microbial profiles of DFM-treated shrimp versus no DFM were significantly different ($P = 0.026$). Shrimp were then used in an early mortality syndrome (EMS) challenge study; EMS is triggered by specific strains of *Vibrio parahaemolyticus* causing acute hepatopancreatic necrosis disease (AHPND). Tanks were stocked with shrimp from both treatments. Two additional tanks were stocked with specific-pathogen-free (SPF) shrimp as an additional control. Shrimp were challenged with AHPND-causing *V. parahaemolyticus* morning and evening on d 0 and 2. Tanks were checked each day for moribund and dead animals. Moribund animals were preserved in fixative to confirm AHPND infection. Resistance to AHPND in DFM-treated animals was 49% versus that of the positive control. Diets supplemented with a DFM can improve growth and survival, modify shrimp digestive microbiome, and improve resistance to pathogenic bacteria (*Vibrio* spp.).

Key words: *Litopenaeus vannamei*, *Bacillus*, *Lactobacillus*

P117 The effect of intra-amniotic and post-hatch dietary synbiotic administration on duodenum *MUC2* gene expression.

A. Calik*[1], F. Dilber[1], P. Sacakli[1], and T. Tekinay[2],
[1]Department of Animal Nutrition Nutritional Diseases, Faculty of Veterinary Medicine, Ankara University, Ankara, Turkey, [2]Life Sciences Research and Application Center, Gazi University, Ankara, Turkey.

The current study was conducted to evaluate the effect of intra-amniotic and post-hatch dietary synbiotic administration on broiler duodenum *MUC2* gene expression. Gene expression in duodenum was assessed on day of hatch and d 42 with RT-PCR technique. In experiment 1, 510 eggs containing viable embryos were divided into 3 groups of 170 eggs each. The first group was not injected and served as a negative control (NC). The second group was injected with 0.9% NaCI and served as positive control (PC). The last group was injected with solutions containing 0.5% inulin and 1×10^6 *Enterococcus faecium* NCIMB 10415 and served as the synbiotic group (S). Hatchlings of non-injected and synbiotic-injected groups were selected for experiment 2. Each group was divided into 2 new groups and birds were offered a basal diet or synbiotic-supplemented diet, as follows; NC-Basal = non-injected birds fed basal diet; S-Basal = synbiotic-injected birds fed basal diet; NC-Syn = non-injected birds fed 1% inulin and 2×10^9 *E. faecium* cfu/kg of feed; S-Syn = synbiotic-injected birds fed 1% inulin and 2×10^9 *E. faecium* cfu/kg feed. Total of 12 hatchlings per group in experiment 1 and 7 birds from each group in experiment 2 were selected and killed for gene expression analysis. Total RNA was isolated using Trizol RNA isolation protocol. Expression of *MUC2* was normalized against an endogenous reference gene (GAPDH) using the comparative threshold cycle (Ct) method to assess the relative mRNA. Gene-specific primers were used for SYBR Green detection according to the published cDNA sequences for each of the studied genes. Our results showed that intra-amniotic synbiotic administration did not affect hatchability or hatching weight of the birds. Duodenum *MUC2* gene expression was increased by intra-amniotic synbiotic administration at day of hatch. However, no differences were observed on d 42. The importance of early-established beneficial microflora in the chicken intestine and its beneficial effect on intestinal health has long been known. Based on the suggested favorable effects of intra-amniotic probiotic or prebiotic administrations, intra-amniotic synbiotic inclusion may be an effective way to maintain the intestinal epithelium integrity of the hatchlings.

P118 Effect of garlic powder and vitamin E-selenium and their combination on performance, immune response, lipid profile, and blood picture of broilers.

Y. J. Jameel*[1,2], M. Hashim[2], M. A. Husain[1], and A. M. Sahib[3],
[1]University of Kerbala, Karbala, Iraq, [2]Texas A&M University, College Station, TX, [3]University of Kufa, Kufa, Najaf, Iraq.

The objective of this study was to evaluate performance, immune response, blood picture, and lipid profile of broilers that were fed diets supplemented with garlic powder (GP; sun-dried) and provided water rich with vitamin E and selenium (Ese; LOVIT E+Se liquid from Lohmann). Three hundred 1-d-old broiler chicks (Ross 308) were randomly assigned to 4 treatment groups: T1 (control), T2 (GP 1%), T3 (ESe 1 mL/L), and T4 (1% GP and ESe 1 mL/L). Each treatments had 3 replicates with 25 birds per replicate. This study used a 3-phase rearing program (16 d starter, 10 d grower, and 9 d finisher) and diets were fed as pellet. Blood samples were collected at the end of the experiment (35 d) to estimate the antibody titer (by ELISA) against ND virus, lipid profile, and blood picture. Body weight and feed consumption were measured at the end of the experiment and the broiler productivity index was calculated. Data were analyzed as one-way ANOVA and significant means separated using Duncan's range test ($P \leq 0.05$). Results revealed that body weight, feed conversion ratio, ELISA antibody titers, total WBC, total RBC, PCV, and Hb, and lipid profile were improved significantly ($P \leq 0.05$) in T4 (chicks supplemented with 1% GP and ESe 1 mL/L) followed by T3 (chicks supplemented with ESe 1mL/liter) and T2 (chicks supplemented with 1% GP) compared with the control group. In conclusion, supplementing broilers diet with 1% GP and drinking water with ESe might be beneficial for broiler performance, immune response, lipid profile, and blood picture of broilers, and the benefits may be due to synergistic effect of garlic and vitamin E-selenium.

P119 Effects of suckling pigs from gilts fed dried bovine plasma during gestation and lactation on intestinal morphometry.

B. V. Freitas*[1], R. A. Nacimento[1], S. M. M. K. Martins[1], L. F. Araújo[1], C. S. S. Araújo[1], and G. Hosotani[2],
[1]University of São Paulo, Pirassununga, SP, Brazil, [2]University of Missouri, Columbia, MO, USA.

The epitheliochorial placenta in pigs does not allow transportation of significant amounts of immunoglobulins from gilts to the fetus, resulting in low immunological resistance neonatal pigs. Therefore, neonatal pigs require passive immunity from colostrum, promoting intestinal immunity by the immunoglobulins. The objective was to evaluate the benefits of plasma inclusion in the diets for gilts on intestinal morphometry of post-weaning piglets.

The experimental design was a factorial arrangement 4 × 2 with 16 gilts randomly assigned to treatments and fed increasing concentrations of plasma (0, 0.5, 1.0, and 2.0%) and 2 collection periods of 5-cm segments from duodenum of 8 piglets at each collection period (d 21 and 61 post-weaning). Samples were processed and analyzed at the Laboratory of Oncology (LOCT-FZEA/USP). Morphometric parameters (villus height, crypt depth, and villus:crypt ratio) were analyzed using SAS software (SAS Institute, Cary, NC) with significance level of 5%. There were no significant differences in villus height, crypt depth, and villus:crypt ratio ($P > 0.05$) from piglets when gilts were fed increasing levels of plasma, and no interactions was observed between plasma levels and period of collection; however, there were significant differences of the effects of collection periods (d 21 or 61) on all morphometric parameters analyzed ($P < 0.05$). These data suggest that feeding plasma to gilts during gestation and lactation does not improve intestinal morphometric parameters in neonatal pigs, and pigs had increased villus height, crypt depth, and decreased villus height:crypt depth at d 61 compared with d 21.

P120 Effect of monocomponent protease on performance, organ size, and duodenal morphology in broilers fed autoclaved soybean.

M. E. Mayorga*[1] and L. B. Moraes[2],
[1]UNIAGRARIA, Bogota, Cundinamarca, Colombia, [2]Institute for Veterinary Research "Desidério Finamor" (IPVDF), Animal Health, State Foundation of Agricultural Research (Fepagro), Eldorado do Sul, Rio Grande do Sul, Brazil.

Raw soybean utilization is limited by the presence of anti-nutritional factors (ANF), which interfere with nutrient digestion and absorption, causing poor performance and leading to damage to the intestinal mucosa. However, they can be partially inactivated through thermal processing and exogenous protease addition. The objective of the current study was to assess the influence of increasing levels of protease on performance, organ weight, and duodenal morphology of broilers fed autoclaved soybean. Four hundred broilers were allocated to 5 dietary treatments for 28 d. Diets were supplemented with 0, 200, 400, 800, and 1,600 ppm protease, and autoclaved soybean was incorporated at inclusion level of 20%. Each treatment had 8 replicates with 10 birds per replicate. Weekly, bird and feed weights were recorded. On d 28, pancreas and duodenum relative weights were calculated. Additionally, on d 14 and 28, duodenum samples were taken for histological measurements. One-way ANOVA was used to analyze the data. On d 14, birds supplemented with 400, 800, and 1,600 ppm of protease exhibited greater body weight gain ($P < 0.005$), and an improvement ($P < 0.005$) in feed conversion ratio (FCR) was observed in supplemented birds regardless of the level of protease. On d 28, 200 and 1,600 ppm of protease improved FCR ($P < 0.005$). Graded inclusion of protease led to a pancreas relative weight reduction (quadratic, $P < 0.0026$). Similarly, 200 and 1,600 ppm of protease elicited smaller duodenum relative weight size (cubic, $P < 0.0162$). On d 14, 200 ppm of protease produced the highest total mucosa thickness and villus height:crypt depth ratio (VCR), and 800 and 1,600 ppm of protease increased crypt depth. On d 28, 400 ppm of protease improved the villus height and total mucosal thickness, and 400 and 800 ppm of protease elicited a higher VCR. In summary, although significant amounts of ANF remained active even after soybean thermal processing, protease supplementation had a beneficial effect on bird performance and enhanced functionality of the intestinal epithelium.

Key words: duodenal morphology, protease, soybean

NOTES

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