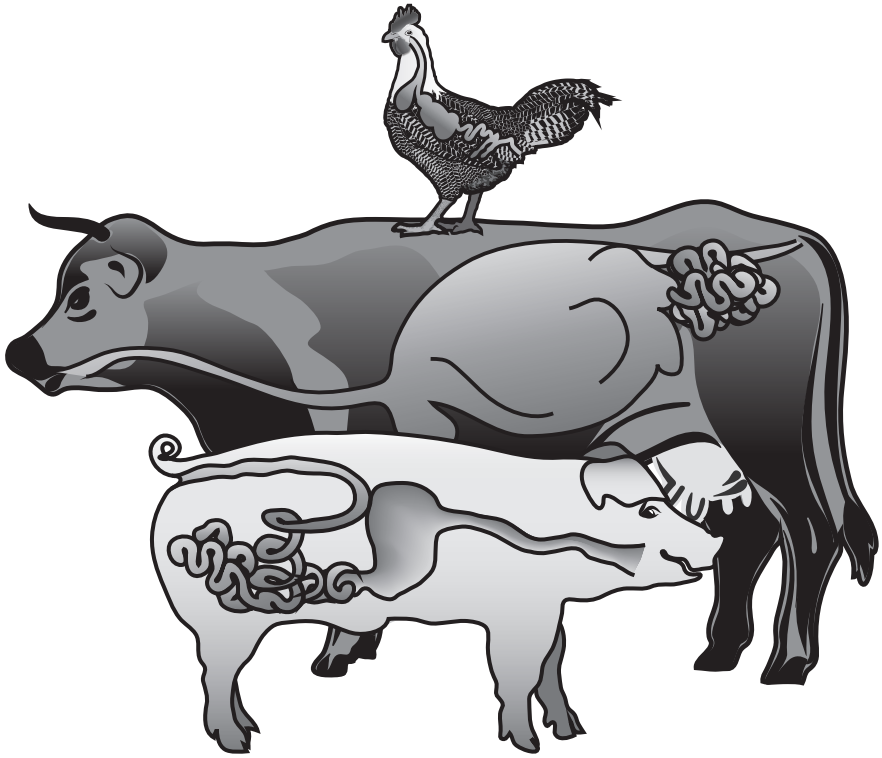
Symposium on Gut Health
in Production of Food Animals
November 10–12, 2014, St. Louis, Missouri



Program and Abstracts
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CONTENTS

WELCOME

On behalf of the Organizing Committee for the 3rd Symposium on Gut Health in Production of Food Animals, I welcome you to St. Louis, Missouri! I hope that the change in venue has made travel to the symposium easier and less expensive.

Like the first two symposia organized around the topic of gut health in food animals, the aim this year 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 feedback that I received from last year's symposium was overwhelmingly positive, as evidenced by the fact that this year, we had twice as many submitted abstracts, covering a broader area of gut health in production animals.

The overall aim of the conference is to promote the unifying concepts that the gut drives animal health and performance. Although the gastrointestinal tract is frequently described simply as "the gut," it is actually made up of (1) an epithelium; (2) a diverse and robust immune arm, which contains most of the immune cells in the body; and (3) the commensal bacteria, which contain more cells than are present in the entire host organism. Understanding of 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 elucidates these links and mechanisms that interconnect the three components of the gut and how each can be manipulated to improve animal health.

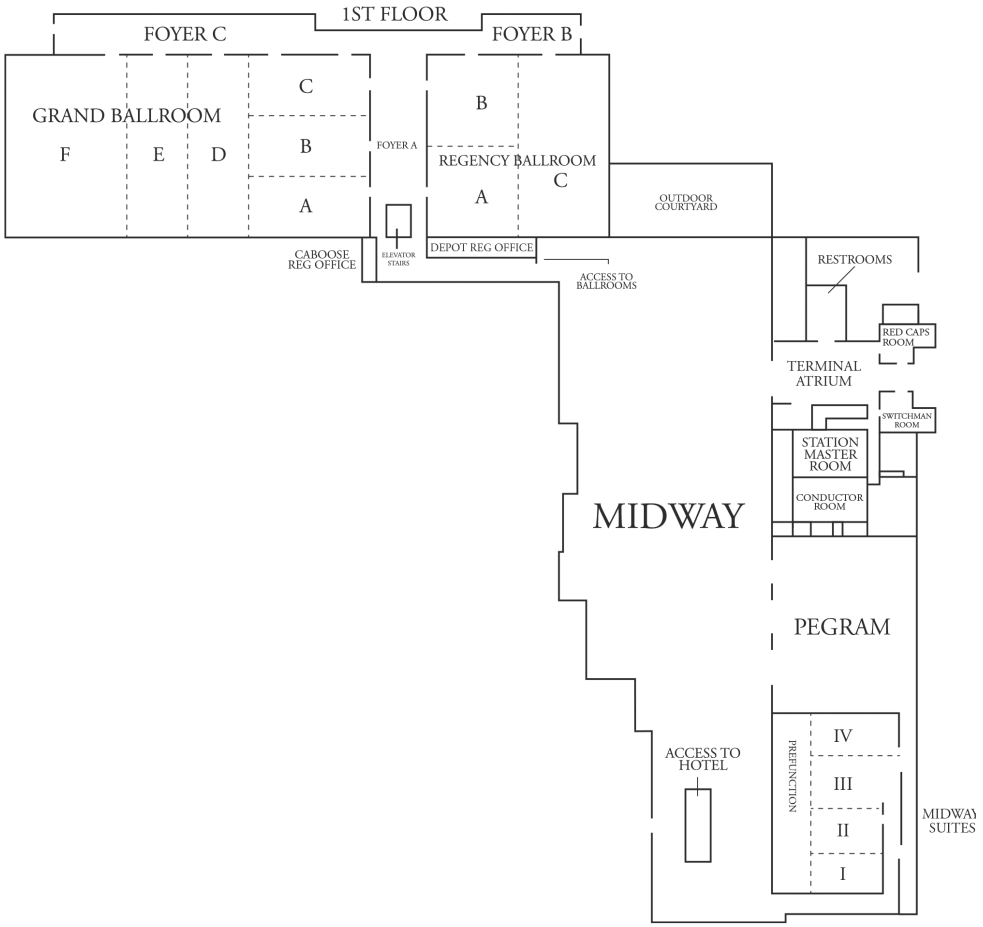
Like last year, we have invited three distinguished plenary speakers, who will cover current research topics in avian, porcine, and bovine gut health. Please take advantage of the presence of these scientists to engage in productive talks and develop collaborations between laboratories in order to further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

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

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

Mike Kogut
Chair, Organizing Committee

St. Louis Union Station Hotel



Program

Sunday, November 9

5:00 pm – 7:00 pm Registration: Foyer B

Monday, November 10

7:00 am – 8:00 am Breakfast: Regency A
Sponsored by King Techina

7:00 am – 12:00 pm Registration: Foyer B

SESSION I: MUCOSAL IMMUNOBIOLOGY/INTESTINAL MUCUS/INTESTINAL BARRIER FUNCTION

Chair: Mike Kogut, USDA-ARS
Regency C

8:00 am Welcome.
Mike Kogut, USDA-ARS, Chair, Organizing Committee.

8:15 am – 9:15 am **Invited presentation:** Role of the microbiome in immune system development in the gastrointestinal tract of newborn calves. (Abstract 100)
P. J. Griebel[*1], N. Malmuthuge[2], G. Liang[2], M. Zhou[2], and L. L. Guan[2], [1]University of Saskatchewan, Saskatoon, SK, Canada, [2]University of Alberta, Edmonton, AB, Canada.*

9:15 am – 9:45 am DIVA defense: Broad protection for *Salmonella* suppression. (Abstract 101)
B. L. Bearson[*1], S. M. D. Bearson[2], I. S. Lee[3], and J. D. Kich[4], [1]USDA, ARS, National Laboratory for Agriculture and the Environment, Ames, IA, USA, [2]USDA, ARS, National Animal Disease Center, Ames, IA, USA, [3]Hannam University, Department of Biological Sciences and Biotechnology, Daejeon, Republic of Korea, [4]Embrapa Swine and Poultry, Concordia, SC, Brazil.*

9:45 am – 10:15 am Identification of potential biomarkers for gut barrier function assay in poultry. (Abstract 102)
J. Chen and J. Escobar, Novus International Inc., St Charles, MO, USA.*

10:15 am – 10:45 am Coffee Break: Foyer B
Sponsored by Chr. Hansen Inc.

10:45 am – 11:15 am Grape seed proanthocyanidins protect the intestinal mucosa barrier from injury induced by weaning stress in piglets. (Abstract 103)
P. Fan, J. Wang, P. Song, D. Li, and X. Ma, State Key Laboratory of Animal Nutrition, Ministry of Agriculture Feed Industry Centre, China Agricultural University, Beijing, China.*

11:15 am – 11:45 am Development of an enteric inflammation model in broilers and methods to detect leaky gut—An overview. (Abstract 104)
L. R. Bielke[*1], V. A. Kuttappan[1], E. A. Vicuña[1], O. B. Faulkner[1], A. D. Wolfenden[1], R. Galarza-Seeber[1], X. Hernandez-Velasco[2], G. Tellez[1], and B. M. Hargis[1], [1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]Facultad de Medicina Veterinaria y Zootecnia, Universidad Nacional Autónoma de México, México.*

11:45 am to 12:15 pm Effects of virginiamycin and/or a xylanase on performance and intestinal health of broiler chickens. (Abstract 105)
H. M. Cervantes[*1,2], M. S. Franca[2], D. T. Elmore[2], G. M. Pesti[2], and K. W. Bafundo[1], [1]Phibro Animal Health, Teaneck, NJ, USA, [2]University of Georgia, Athens, GA, USA.*

12:15 pm – 1:15 pm Lunch: Regency A

1:15 pm – 3:15 pm Poster Session: Foyer A and Foyer B

SESSION II: BENEFICIAL MICROBES AND GUT HEALTH

Chair: Mike Kogut, USDA-ARS
Regency C

3:15 pm – 4:15 pm **Invited presentation:** Microbiome modulation in turkeys: Friend or foe? (Abstract 106)
T. J. Johnson, University of Minnesota, Saint Paul, MN.*

4:15 pm – 4:45 pm Effect of *Megasphaera elsdenii* NCIMB 41125 dosing on rumen development, volatile fatty acid production and blood β-hydroxybutyrate in neonatal dairy calves. (Abstract 107)
M. C. Muya[*1], F. V. Nherera[1], K. A. Miller[2], C. C. Aperce[2], and L. J. Erasmus[3], [1]Agricultural Research Council, Pretoria, South Africa, [2]MS Biotec, Wamego, KS, USA, [3]University of Pretoria, Pretoria, South Africa.*

4:45 pm – 5:15 pm Prebiotics and probiotics reduce enterotoxigenic *Escherichia coli* and *Salmonella enterica* infectivity in an in vitro bovine cell model. (Abstract 108)
D. Baines[*1] and R. Lowe[2], [1]Lethbridge Research Centre, Lethbridge, Alberta, Canada, [2]University of Alberta, Edmonton, Alberta, Canada.*

5:15 pm – 5:45 pm *Bacillus amyloliquefaciens* CECT 5940 persistency test in chicken gut. (Abstract 109)
A. Ortiz, P. Honrubia, and J. J. Mallo, NOREL S.A., Madrid, Spain.*

5:45 pm – 6:15 pm Evaluation of a novel probiotic formulation designed to reduce decreased body weight gain in chicks following vaccination with commercially available coccidiosis vaccines. (Abstract 110)
M. F. Faulkner, J. D. Lum, J. L. Vicente, and R. E. Wolfenden, Pacific Vet Group, Farmington, AR, USA.*

6:15 pm – 6:45 pm Evaluation of probiotic and prebiotic effects on dairy cattle performance. (Abstract 111)
D. Baines, Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada.*

7:00 pm – 9:00 pm Reception: Regency C
Sponsored by Cobb-Vantress Inc.

Tuesday, November 11

SESSION III: BENEFICIAL MICROBES AND GUT HEALTH (CONTINUED)

Chair: Mike Kogut, USDA-ARS
Regency C

7:00 am – 8:00 am Breakfast: Regency A

8:00 am – 9:00 am **Invited presentation:** Breaking down barriers: The impact of production stressors on gut immune and epithelial defense in the pig. (Abstract 200)
A. Moeser, North Carolina State University, Raleigh, NC, USA.*

9:00 am – 9:30 am A late finishing *Lawsonia intracellularis* challenge is moderated by Lincomix feed medication. (Abstract 201)
D. A. Nelson[*1], P. Knoernschild[1], R. Fleck[1], N. Winkelman[2], A. Mueller[2], and D. Amodie[1], [1]Zoetis Inc., Florham Park, NJ, USA, [2]Swine Services Unlimited Inc., Rice, MN, USA.*

9:30 am – 10:00 am Evaluation of prebiotics and probiotics to reduce toxicity of pure and mixed-feed mycotoxins in vitro and to prevent carry-over of aflatoxin B1 in dairy cows. (Abstract 202)
D. Baines, Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada.*

10:00 am – 10:30 am Coffee Break: Foyer B
Sponsored by Lesaffre

10:30 am – 11:00 am Effects of *Lactobacillus fermentum* I5007 supplementation on lipogenesis and adipose tissue distribution. (Abstract 203)
J. Chen, L. Li, X. Ma, S. Qiao, and D. Li, State Key Laboratory of Animal Nutrition, Ministry of Agriculture Feed Industry Centre, China Agricultural University, Beijing, China.*

11:00 am – 11:30 am Evaluation of the efficacy of *Enterococcus faecium* CECT4515 and *Bacillus licheniformis* to improve piglet performance. (Abstract 204)
A. Ortiz[*1], G. Cano[2], P. Honrubia[1], and J. J. Mallo[1], [1]NOREL, S.A., Madrid, Spain, [2]Test and Trials, S.L., Huesca, Spain.*

11:30 am – 12:00 pm Comprehensive study of swine and poultry probiotics showed high variation in quality and mode of action. (Abstract 205)
B. K. K. Nielsen, J. M. Nielsen, T. Styrishave, D. Sandvang, and O. R. Sjoeholm, Chr. Hansen A/S, Hørsholm, Denmark.*

12:00 pm – 1:30 pm Lunch: Regency A

1:30 pm – 2:00 pm Applications of a probiotic and prebiotic alleviate polymicrobial mastitis in dairy cattle. (Abstract 206)
D. Baines[*1], J. Zlosnik[2], D. Speert[2], M. Mulvey[3], and L. Masson[4], [1]Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, [2]University of British Columbia, Vancouver, BC, Canada, [3]National Microbiology Laboratory, Winnipeg, MB, Canada, [4]NRC-Montreal, Montreal, QC, Canada.*

2:00 pm – 2:30 pm Applications of a probiotic and prebiotic alleviate mammary aspergillosis in dairy cattle. (Abstract 207)
D. Baines, Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada.*

2:30 pm – 2:45 pm The role of a selected *Bacillus subtilis* direct-fed microbial in performance, intestinal viscosity, bacterial translocation, and bone mineralization in broiler chickens fed with high-NSP diets. (Abstract 208)
J. D. Latorre[*1], V. A. Kuttappan[1], M. H. Kogut[2], A. Wolfenden[1], X. Hernandez-Velasco[3], L. R. Bielke[1], B. M. Hargis[1], O. B. Faulkner[1], and G. Tellez[1], [1]University of Arkansas, Department of Poultry Science, Fayetteville, AR, USA, [2]USDA-ARS, SPARC, College Station, TX, USA, [3]Universidad Nacional Autonoma de Mexico, Mexico.*

2:45 pm – 3:15 pm Molecular survey of the microbiomes in broilers to understand probiotics-induced reduction of BCO lameness. (Abstract 209)
T. Jiang[1], R. K. Mandal[1], R. F. Wideman Jr.[1], T. Lohrmann[2], and Y. M. Kwon[*1], [1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]Quality Technology International Inc., Elgin, IL, USA.*

3:15 pm to 3:45 pm Coffee Break: Foyer B

SESSION IV: DIVERSITY OF THE MICROBIOME/IMPACT OF GUT MICROBIAL COMMUNITIES

Chair: Mike Kogut, USDA-ARS
Regency C

3:45 pm – 4:15 pm — Development of the microbiome of chicks: Early exposure influences future microbial diversity, independent of colonization. (Abstract 210)
A. Ballou, R. Ali, M. Mendoza, H. Hassan, and M. Koci, North Carolina State University, Raleigh, NC, USA.*

4:15 pm – 4:45 pm — Transfer of heifers calves from maternity to calf pen: The impact on the population of enterobacteria. (Abstract 211)
J. F. Reis, S. M. F. Novo, C. C. Baccili, C. R. Stricagnolo, B. T. Silva, and V. Gomes, University of São Paulo, São Paulo, Brazil.*

4:45 pm – 5:15 pm — *Eimeria maxima* causes changes in mRNA expression of genes associated with amino acid and sugar uptake in the jejunum of infected broilers. (Abstract 212)
K. Miska and R. Fetterer, USDA/ARS, Beltsville, MD, USA.*

5:15 pm – 5:45 pm — Interrelationships of fungal and bacteria populations within the gastrointestinal tract of poultry. (Abstract 213)
J. A. Byrd, USDA-ARS, Food and Feed Safety Research Unit, College Station, TX, USA.*

5:45 pm – 6:15 pm — Modification of the chicken cecal microbiome by *Campylobacter jejuni* colonization and by a feed additive. (Abstract 214)
A. Thibodeau[*1], P. Fravalo[1], E. Yergeau[2], L. Lahaye[3], J. Arsenault[1], and A. Letellier[1], [1]Université de Montréal, Saint-Hyacinthe, QC, Canada, [2]Conseil national de recherche du Canada, Montréal, QC, Canada, [3]Jefo, Saint-Hyacinthe, QC,Canada.*

7:00 pm – 9:00 pm — Reception: Regency C
Sponsored by Cargill Animal Nutrition

Wednesday, November 12

SESSION V: NUTRITION AND GUT HEALTH

Chair: Mike Kogut, USDA-ARS
Regency C

7:00 am – 8:00 am — Breakfast: Regency A

8:00 am – 8:30 am — Impact of a multistrain *Bacillus* product on broiler performance and small intestinal microbiota. (Abstract 300)
M. Hruby[*1], J. C. Remus[1], and A. J. Madisen[2], [1]Danisco Animal Nutrition, DuPont Industrial Biosciences, St. Louis, MO, USA, [2]Animal & Environmental Application, DuPont Nutrition & Health, Waukesha, WI, USA.*

8:30 am – 9:00 am — Utilization of rye as energy source affects bacterial translocation, intestinal viscosity, microbiota composition, and bone mineralization in broiler. (Abstract 301)
G. Tellez[*1], J. D. Latorre[1], V. Kuttappan[1], A. Wolfenden[1], M. Kogut[2], X. Hernandez-Velasco[3], B. M. Hargis[1], O. Faulkner[1], W. Bottje[1], and L. R. Bielke[1], [1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]USDA-ARS, SPARC, College Station, TX, USA, [3]Facultad de Medicina Veterinaria y Zootecnia, Universidad Nacional Autónoma de México, México.*

9:00 am – 9:30 am Impact of dietary exogenous enzyme supplementation on enteric adherent mucin thickness layer and gastrointestinal morphological development in poultry. (Abstract 302)
A. A. Ayoola, P. R. Ferket, R. D. Malheiros, and J. Grimes, Prestage Dept. Poultry Science, NCSU, Raleigh, NC, USA.*

9:30 am – 10:00 am Phytogenic feed additives as replacement for antibiotic growth promoters in broiler chickens. (Abstract 303)
G. R. Murugesan[1], B. Syed[2], and S. Haldar[3], [1]Biomin America Inc., San Antonio, TX, USA, [2]Biomin Holding GmbH, Herzogenburg, Austria, [3]Department of Animal Nutrition, West Bengal University of Animal & Fishery Sciences, Kolkata, India.*

10:00 am – 10:30 am Coffee Break: Foyer B

10:30 am – 10:45 am Gut health model using different diet formulations in broiler chickens. (Abstract 304)
V. A. Kuttappan[1], J. D. Latorre[1], K. Wedekind[2], J. Escobar[2], E. A. Vicuña[1], R. Galarza[1], O. B. Faulkner[1], A. D. Wolfenden[1], G. I. Tellez[1], B. M. Hargis[1], M. Vazquez-Anon[2], and L. R. Bielke[1], [1]Department of Poultry Science, University of Arkansas, Fayetteville, AR, USA, [2]Novus International, Inc., St. Charles, MO, USA.*

10:45 am – 11:15 am Protein-mediated butyrate transport in the rumen epithelium is modulated by feed restriction in Holstein steers. (Abstract 305)
A. H. Laarman[1], R. A. Pederzolli[2], G. B. Penner[2], and B. W. McBride[1], [1]University of Guelph, Guelph, ON, Canada, [2]University of Saskatchewan, Saskatoon, SK, Canada.*

11:15 am – 11:45 am Effects of Sporulin and Cibenza CSM on gut health and growth performance of broilers. (Abstract 306)
J. Chen, K. J. Wedekind, and J. J. Dibner, Novus International Inc., St Charles, MO, USA.*

Poster Presentations

P100 The effectiveness of direct-fed microbial and prebiotic on histomorphology of intestine, ultrastructural changes of intestinal mucosa and performance of turkey poults infected with *Salmonella* and *Campylobacter*.
S. Rahimi[*1], J. Grimes[2], S. Kathariou[2], and O. Fletcher[2], [1]Tarbiat Modares University, Tehran, Tehran, Iran, [2]North Carolina State University, Raleigh, NC, USA.*

P101 A review: Alternatives to antibiotic use for growth promotion in poultry.
S. Rahimi[*1], M. Naghizadeh[1], and A. Rahimi[2], [1]Tarbiat Modares University, Tehran, Tehran, Iran, [2]Islamic Azad University, Tehran, Tehran, Iran.*

P102 Selective isolation of gut lactic acid bacteria from commercial beef cattle originating from farms at risk of subacute ruminal acidosis (SARA).
R. C. Cernat[*1], C. A. McCartney[1], I. Hindrichsen[2], E. Brockmann[2], C. H. H. Koh-Tan[3], E. M. Strachan[4], W. Thomson[4], T. J. Snelling[1], C. D. Harvey[4], N. N. Jonsson[3], and J. R. Wallace[1], [1]Rowett Institute of Nutrition and Health, University of Aberdeen, Aberdeen, UK, [2]Chr. Hansen A/S, Hørsholm, Denmark, [3]Animal Health and Comparative Medicine, Institute of Biodiversity, University of Glasgow, Glasgow, UK, [4]Harbro Ltd., Turriff, UK.*

P103 Dynamics of the microbiome over the rearing period in two lines of broilers.
S. Diaz Sánchez[*1], R. Hawkins[2], R. Okimoto[2], A. Layton[3], A. Saxton[4,5], J. A. Blakeley-Ruiz[5], and I. Hanning[1,5], [1]Department of Food Science and Technology, University of Tennessee, Knoxville, TN, USA, [2]Cobb-Vantress Incorporated, Siloam Springs, AR, USA, [3]Department of Microbiology, University of Tennessee, Knoxville, TN, USA, [4]Animal Science, University of Tennessee, Knoxville, TN, USA, [5]Department of Genome Sciences and Technology, University of Tennessee, Knoxville, TN, USA.*

P104 Neonatal lambs' gastrointestinal tracts are initially colonized by a unique and dynamic vaginal microbiota but rapidly transition toward the dams' teat.
M. Lachman, J. Swartz, K. Westveer, T. O'Neill, J. B. Geddes, and C. J. Yeoman, Montana State University, Bozeman, MT, USA.*

P105 Variations of the microbiome among sheep breeds on two different diets.
T. M. Taxis[*1], M. J. Ellison[2], K. M. Cammack[2], G. C. Conant[1], and W. R. Lamberson[1], [1]University of Missouri, Columbia, MO, USA, [2]University of Wyoming, Laramie, WY, USA.*

P106 A calf-rearing model for exploring the influence of colostrum on the microbiological health of the developing bovine intestinal tract.
E. B. Bichi[*1], C. J. Yeoman[1], J. Lowe[1], N. Maradiaga[1], E. Pulido Chavez[3], and B. Aldridge[1], [1]Integrated Food Animal Systems, College of Veterinary Medicine, University of Illinois, Urbana-Champaign, IL, USA, [2]Department of Animal and Range Sciences, Montana State University, Bozeman, MT, USA, [3]Instituto De Ganaderia De Montana, Leon, Spain.*

P107 Genetic and genome analyses of bacteria cultured from lame broilers with osteomyelitis.
A. A. K. Al-Rubaye, D. D. Rhoads, and R. F. Wideman, University of Arkansas, Fayetteville, AR, USA.*

P108 Proteins involved in intracellular pH regulation in rumen epithelial cells are modulated during the transition period in Holstein dairy cows.
A. H. Laarman[*1], A. Kleinberg[1], M. A. Steele[2], O. AlZahal[1], and B. W. McBride[1], [1]University of Guelph, Guelph, ON, Canada, [2]University of Alberta, Edmonton, AB, Canada.*

P109 High and low loads of cecal colonization by *Salmonella* Enteritidis in chickens triggers distinct immune kinome profiles.
C. L. Swaggerty, R. J. Arsenault, and M. H. Kogut, USDA/ARS, College Station, TX, USA.*

P110 Norepinephrine modulates swine gut immune cells.
E. Silva, D. Lay, and S. Eicher, USDA-ARS-MWA Livestock Behavior Research Unit, West Lafayette, IN, USA.*

P111 A role for the noncanonical Wnt-signaling pathway in the induction of a state of immune tolerance that allows the establishment of persistent intestinal colonization of *Salmonella enterica* serovar Enteritidis in chickens.
M. H. Kogut, C. L. Swaggerty, K. J. Genovese, H. He, and R. J. Arsenault, USDA-ARS, SPARC, College Station, TX, USA.*

P112 Anti-interleukin-10 antibody is effective at eliminating the adverse effects of a coccidiosis challenge.
J. Sand, A. Repasy, J. Roberts, and M. Cook, University of Wisconsin, Madison, WI, USA.*

P113 Comparison of anti-interleukin-10 egg antibody to Maxiban in coccidia-infected broiler chicks.
M. Arendt[1], J. Sand[2], and M. Cook[2], [1]Department of Comparative Biosciences, University of Wisconsin-Madison, Madison, WI, USA, [2]Department of Animal Science, University of Wisconsin-Madison, Madison, WI, USA.*

P114 Effects of dietary fiber on cecal short-chain fatty acid and microbial community of broiler and layer chicks.
M. Walugembe, J. Hsieh, N. Koszewski, S. Lamont, M. Rothschild, and M. Persia, Iowa State University, Ames, IA, USA.*

P115 Effects of *Saccharomyces cerevisiae* fermentation products on fiber digesting and lactate utilizing rumen bacteria at neutral and low pH in vitro.
A. Brainard, V. Nsereko, I. Yoon, J. Butler, and M. Scott, Diamond V, Cedar Rapids, IA, USA.*

P116 Synergistic effect of *Bacillus licheniformis* and Flavomycin on broiler performance.
A. B. Kehlet[1], H. Kling[3], M. Sims[2], and D. Harrington[1], [1]Chr. Hansen A/S, Hørsholm, Denmark, [2]Virgina Diversified Research Corp., Harrisonburg, VA, USA, [3]Chr. Hansen, Milwaukee, WI, USA.*

P117 Effect of quercetin on performance, apparent digestibility of feed nutrients. and cecal microbiota in laying hens at 39 to 47 weeks old.
Y. Li, N. Teng, M. T. Chaudhry, C. Y. Han, D. T. Sun, Y. You, and L. Li, Institution of Animal Nutrition, Harbin, Heilongjiang, China.*

Session I: Mucosal Immunobiology/Intestinal Mucus/ Intestinal Barrier Function

100 Role of the microbiome in immune system development in the gastrointestinal tract of newborn calves.

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

There is increasing evidence that the commensal microbiome has diverse effects on mucosal immune system development and function. In newborn calves, it is a challenge to clearly delineate the effects of the microbiome from other contributing factors, such as diet, environment, and host genetics. The bovine gastrointestinal tract (GIT) is rapidly colonized during birth and during the neonatal period a succession of microbial species contributes to increased microbial density and diversity. This succession occurs rapidly during the first week of life and then progresses much more slowly. Characterization of the microbiome in the neonatal GIT revealed marked bacterial variation, at both family and species level, among individual animals. The microbiome composition also varied significantly when comparing ingesta- and mucosa-associated communities within individual GIT regions. The first week postpartum is a very dynamic developmental period in the bovine GIT with significant changes in both mucosal barrier and immune function. Differential expression (DE) of intestinal miRNAs confirms that the greatest change in GIT development occurred during the first week of life with DE miRNAs involved in regulating both GIT developmental and immunological processes. Relatively few miRNAs were differentially expressed when comparing tissues collected from 6-wk-old calves and 3-wk-old calves. Correlation analyses of total bacterial numbers and specific families revealed significant associations between the commensal microbiome and the expression of genes involved in regulating both mucosal barrier and innate immune function. This analysis also suggests that regional differences in the microbiome may be associated with significant regional differences in the expression of innate immune genes. This information provides the baseline to begin analyzing the role of individual bacterial species or microbial communities in shaping early mucosal immune system development and the long-term health of calves.

Key Words: bovine, gastrointestinal tract, mucosal immunity microbiome, microRNA

101 DIVA defense: Broad protection for *Salmonella* suppression.

B. L. Bearson*[1], S. M. D. Bearson[2], I. S. Lee[3], and J. D. Kich[4],
[1]USDA, ARS, National Laboratory for Agriculture and the Environment, Ames, IA, USA, [2]USDA, ARS, National Animal Disease Center, Ames, IA, USA, [3]Hannam University, Department of Biological Sciences and Biotechnology, Daejeon, Republic of Korea, [4]Embrapa Swine and Poultry, Concordia, SC, Brazil.

A live, attenuated *Salmonella enterica* serovar Typhimurium vaccine was developed to confer broad protection against multiple *Salmonella* serovars to prevent disease and reduce pathogen colonization and shedding. Two vaccine trials were performed in swine to determine the protection afforded by the vaccine to challenge with 2 distinct *Salmonella* serovars. In the first vaccine trial, pigs were administered 2 doses of the vaccine (vaccination and booster) and challenged with wild-type *Salmonella* Typhimurium UK1 that causes gastroenteritis. The swine rectal temperatures and plasma IFNγ levels were significantly reduced in vaccinated pigs compared with mock-vaccinated swine. Fecal shedding and tissue colonization with wild-type *Salmonella* Typhimurium UK1 was significantly reduced in vaccinated pigs compared to mock-vaccinated swine. Furthermore, the vaccine strain did not induce a serological response to *Salmonella* LPS; therefore, an ELISA can be used to differentiate infected from vaccinated animals (DIVA). In the second vaccine trial, pigs were administered a single dose of the vaccine and challenged with a virulent *Salmonella* Choleraesuis that causes systemic disease in swine. Compared with the mock-vaccinated group, the vaccinated pigs exhibited significantly reduced rectal temperatures, serum IFNγ levels, and tissue colonization. Furthermore, during the challenge period, the isolation of *Salmonella* Choleraesuis from blood cultures was significantly greater in mock-vaccinated pigs compared with vaccinated swine. The data from these 2 vaccine trials indicate that the live, attenuated *Salmonella* Typhimurium vaccine can both protect swine from *Salmonella* that cause systemic disease in pigs and also reduce the potential for transmission of *Salmonella* that cause foodborne disease in humans. Therefore, this dual-use *Salmonella* DIVA vaccine is intended to not only protect the health status of the swine herd to reduce production losses, but also support food safety and public health by reducing the spread of human foodborne *Salmonella* from pen to plate.

Key Words: *Salmonella*, vaccine, DIVA, swine

102 Identification of potential biomarkers for gut barrier function assay in poultry.

J. Chen* and J. Escobar,
Novus International Inc., St Charles, MO, USA.

Poor gut health can negatively affect performance, reduce nutrient absorption, increase feed passage leading to diarrhea, result in wet or caked litter, reduce uniformity, and increase mortality. One critical aspect of gut heath is barrier function. Gut epithelium with its tight junctions, mucin layers, and gut-associated immune tissue protects animals against invasion by opportunistic pathogens from the microbiota. The objective of this trial was to identify biomarkers to measure gut barrier function. A total of 144 day-of-hatch Ross 308 male broiler chicks were housed in battery cages with 6 chicks per cage and fed a common starter diet during d 0–14. On d 14, birds were randomly assigned to 2 treatments: control (CON) and gut barrier failure (GBF). CON birds were fed corn-soybean meal-based diet whereas GBF birds were fed rye-wheat-barley-based diet, housed in cages with floors covered with paper, and orally challenged with coccidiosis vaccine at d 21 to ensure mild gut barrier failure. Birds were euthanized on d 28 to measure gut health parameters. GBF birds had lower ($P < 0.0001$) growth performance than CON birds. Gut morphometry results showed that, compared with CON birds,

GBF birds had greater villus width, crypt depth and crypt/villus ratio and obvious structural damage on villi tips in duodenum, as well as greater villus height, crypt depth and crypt/villus ratio in jejunum indicating that gut barrier failure was occurring in GBF birds. Serum endotoxin levels were increased ($P < 0.0001$) by 2-fold in GBF birds compared with CON birds. The mRNA levels of gut barrier-associated genes in jejunal mucosa were measured by qRT-PCR. GBF birds had lower levels of fatty acid-binding protein 2 (FABP2, $P = 0.005$), MUC2 ($P = 0.09$) and occludin ($P = 0.10$), a 4-fold increase of FABP6 ($P = 0.02$) and a 3-fold increase of interleukin-8 (IL-8, $P < 0.0001$), compared with CON birds. These results suggest that serum endotoxin and gene expression of *FABP2*, *FABP6*, *IL8*, *MUC2*, and occludin in mucosa are potential biomarkers of gut barrier health.

Key Words: gut barrier, biomarker, endotoxin, gene expression

103 Grape seed proanthocyanidins protect the intestinal mucosa barrier from injury induced by weaning stress in piglets.

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The present study was conducted to determine the effects of grape-seed procyanidins (GSP) on growth performance, hematological parameters, intestinal barriers and antioxidant capacity, as well as gut microbiota in weaned piglets. According to their body weight, litters and sex, 120 crossbreed weaned piglets (Duroc × Landrace × Large White) were assigned to 4 treatments: Control group, GSP group, Antibiotic group, GSPs + Antibiotic group. At d 29, intestinal permeability was detected and the experiments were terminated to obtain the intestinal sample and digesta of piglets. The results showed that GSPs significantly improved the average daily gain (ADG; $P < 0.05$) and feed/gain (F/G; $P < 0.05$) of piglets, respectively compared with the control group, meanwhile the average daily feed intake (ADFI) has no significant difference between other 2 groups. Compared with the control group, the incidence of diarrhea was decreased ($P < 0.05$) in the GSP group and Antibiotic group. The GSPs significantly reduced the urinary lactulose to mannitol ratio of piglets, compared with control groups ($P < 0.05$), which indicated that GSPs decreased the intestinal permeability. The levels of SOD ($P < 0.05$), GSH ($P < 0.05$), and GSH-Px ($P < 0.05$) in serum and intestinal samples were significantly increased in the GSPs group, and the level of MDA was significantly decreased ($P < 0.05$). The concentration of acetic acid, propionic acid, and butyric acid in the colon in the GSPs group and in the antibiotic group were lower than the control group ($P < 0.05$); meanwhile, there was no difference between the GSPs group and the antibiotic group. GSPs probably inhibited bowel microbial fermentation or promoted intestinal epithelial cell to absorb bowel short-chain fatty acids. The distributions and species change of gut microbiota after feeding GSPs requires further study. In conclusion, supplementation with a low dose of GSPs in diets can increase the ADG and F/G ratio while leading to improved growth performance and diarrhea incidence of piglets, with no side effects being observed.

Key Words: grape-seed procyanidins, weaning stress, intestinal permeability, mucosa barrier

104 Development of an enteric inflammation model in broilers and methods to detect leaky gut—An overview.

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One mechanism by which antibiotic growth promoters increase performance parameters in poultry may be through control of enteric inflammation. With the decreasing acceptance of growth promoters in poultry, alternatives must be sought to sustain growth and health of flocks, and research models capable of inducing and measuring changes in enteric inflammation are urgently needed. Multiple induction models and markers of reduced enteric integrity have been investigated. Markers of inflammation such as fluorescein isothiocyanate dextran (FITC-D; 3–5kDa), xanthophyll absorption, bacterial translocation (BT) of Salmonella, and serum opacity have been investigated as markers of decreased enteric integrity. While some of the markers have shown promise, FITC-D has proven consistent and reliable for detecting tight junction leakage, and optimization experiments have investigated parameters such as molecule size and dose. Furthermore, induction methods have included dextran sodium sulfate (DSS), feed restriction (FR), high fat diet, rye-based diet, and oral dexamethasone. While DSS clearly caused intestinal inflammation, lesions were mostly limited to the cecum and treatment effects were generally noted only when severe illness was induced. Presently, FR and rye-based diet have consistently resulted in increased tight junction leakage ($P < 0.05$), though oral dexamethasone has shown promise with increased serum FITC-D and bacterial translocation ($P < 0.05$) in only a single experiment. These enteric inflammation research models provide a means by which alternative antibiotic growth promoters and interventions after intestinal insult can be investigated. Additionally, there is interest in determining if enteric inflammation markers can predict flock health and well-being.

Key Words: enteric inflammation, FITC-D, tight junctions, bacterial translocation

105 Effects of virginiamycin and/or a xylanase on performance and intestinal health of broiler chickens.

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A 44-d floor pen experiment was conducted with 960 Cobb-500 male broiler chickens to investigate the individual and combined effects of 20 ppm dietary virginiamycin (VM) and 10,880 BXU/kg dietary xylanase (XL) and its potential interactions on performance and intestinal health. A 2 × 2 factorial experiment with 12 replicates of 20 birds each per treatment was carried out in a complete randomized block design with 2 dietary concentrations of VM (0 or 20 ppm) and 2 dietary concentrations of XL (0 or 10,880 BXU/kg). Chickens and feeds were weighed at 14, 35 and 44 d to calculate average body weight (BW) and feed conversion ratio (FCR) per treatment. ANOVA was conducted on all data to determine if there were significant (P <

0.05) differences between treatments or the main effect means. To investigate the effect of treatments on intestinal health, 3 sections of the intestine (duodenum, jejunum and ileum) from 12 birds from each treatment (1 bird per replicate) were collected on d 44 while the birds were still on full feed and also after the birds had been fasted for 10 h. The intestinal sections were collected immediately after euthanasia and they were placed in 10% NBF and subsequently cut and stained for histological examination. Inflammatory changes, length of villi, depth of crypts and other morphometric measurements were evaluated by an ACVP board-certified poultry pathologist. On d 0 no differences were detected in BW ($P > 0.9381$), on d 14, 35 and 44 VM supplementation produced significantly heavier BW ($P < 0.05$) and significantly improved FCR ($P < 0.05$). XL supplementation did not have a significant effect at any period, however, a significant interaction ($P < 0.0041$) between VM and XL was found for FCR at 44 d in which the best FCR was attained when both supplements were added to the diet. Results from the histological examination of intestinal sections will be included in the presentation.

Key Words: virginiamycin, xylanase, intestinal health, broiler performance

Session II: Beneficial Microbes and Gut Health

106 Microbiome modulation in turkeys: Friend or foe?

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The microbial communities of an animal (the microbiome) play an essential role in aiding in the development of a healthy gut enabling effective growth and development. There are numerous approaches to modulate the gut microbiome, including subtherapeutic antibiotics, prebiotics, and probiotics. Numerous commercial direct-fed microbial products are available with claims of healthy gut development; unfortunately, the number of products available and different approaches used to test them make it difficult for a producer to determine the optimal approach. In this presentation, a systematic approach used in commercial turkeys toward the identification of optimal bacteria for modulating the gut will be highlighted. This approach is focused upon initial understanding of the baseline succession of the microbiome in the gut, followed by assessment of various combinations of bacteria toward modulating the gut early in life. The key aspect of this approach is the identification of host-specific and time-specific combinations of bacteria that will be most effective across a broad array of management situations. This approach holds promise for the development and refinement of products that can supplant antimicrobials in a reproducible and cost effective manner.

107 Effect of *Megasphaera elsdenii* NCIMB 41125 dosing on rumen development, volatile fatty acid production and blood β-hydroxybutyrate in neonatal dairy calves.

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Thirty calves were randomly assigned to 2 treatments and fed until weaning at 42 d of age. Treatments were a control group, which did not receive *Megasphaera elsdenii* (Me0) and a *M. elsdenii* group, which received a 50-mL oral dose of *M. elsdenii* NCIMB 41125 (10^8 cfu/mL) at 14 d of age (Me14). Calves were given colostrum for the first 3 d of life followed by limited whole milk feeding. A commercial calf starter was offered ad libitum starting at 4 d of age until the end of the study. Fresh water was available throughout the study. Feed intake and growth were determined. Blood samples were collected via jugular venipuncture to determine β-hydroxybutyrate (BHBA) concentrations. Fourteen male calves (7 per group) were euthanized on d 42 and digestive tracts harvested. Reticulo-rumen weight was determined and rumen tissue samples collected from the cranial and caudal sacs of the ventral and dorsal portions of the rumen for measurements of papillae length, papillae width, and rumen wall thickness. Dosing with *M. elsdenii* NCIMB 41125 improved starter DMI ($P < 0.01$), weaning BW ($P < 0.03$), and tended to improve average daily gain ($P = 0.10$). Me14-calves had greater plasma BHBA concentration than Me0-calves during the last 3 wk of the trial ($P < 0.003$). Me14 had greater reticulo-rumen weight ($P = 0.01$), papillae width ($P < 0.001$) and papillae density ($P = 0.02$) compared with Me0. No differences in rumen wall thickness or papillae length were observed between the 2 groups ($P > 0.05$). Total VFA, acetate, and propionate did not differ between treatments ($P > 0.05$), but butyrate concentration was greater in Me14 than Me0 ($P < 0.04$). Results suggest increased butyrate in the presents of *M. elsdenii* increased epithelium metabolism and ketogenesis resulting in greater absorptive area and improved absorption of digestive end products.

Key Words: *Megasphaera elsdenii*, rumen development, neonatal calves, weaning

108 Prebiotics and probiotics reduce enterotoxigenic *Escherichia coli* and *Salmonella enterica* infectivity in an in vitro bovine cell model.

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Enterotoxigenic *Escherichia coli* (ETEC; F4, F5, F18, F41) and *Salmonella enterica* (serovars Dublin, Typhimurium, Anatum, Newport) infections cause morbidity and mortality losses in cattle production. Antimicrobial activities of a probiotic and prebiotic in vitro have recently been validated as novel tools to effectively eliminate Shiga-toxin producing *E. coli* (STEC) infections in all age classes of cattle. The present study aims to evaluate the antimicrobial activities of commercial probiotics (BioSaf, Dairyman's Choice, MaxiPlex) and prebiotics (Celmanax, Safmannan, Bio-Mos, Integral), in preventing *S. enterica* and ETEC colonization of a bovine colonic cell line in vitro. There was a significant dose-dependent reduction in ETEC strain colonization in the presence of Celmanax, Safmannan, Biosaf and Dairyman's Choice, but not in the presence of MaxiPlex, Bio-Mos and Integral ($P = 0.001$). There was a significant dose-dependent reduction in *S. enterica* serovar colonization in the presence of Celmanax, Safmannan, Biosaf and Dairyman's Choice, but not in the presence of MaxiPlex, Bio-Mos, and Integral ($P = 0.001$). Overall, our results suggested that commercial prebiotics and probiotics vary greatly in terms of their antimicrobial activities, but as for studies with STEC, Celmanax and Dairyman's Choice function at dosages achievable in commercial feeds and have a broader spectrum of antimicrobial activities than other commercial formulations.

Key Words: enterotoxigenic *Escherichia coli*, *Salmonella enterica*, prebiotic, probiotic, antimicrobial

109 *Bacillus amyloliquefaciens* CECT 5940 persistency test in chicken gut.

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The objective of the present study was to demonstrate that the DFM *Bacillus amyloliquefaciens* is capable of remaining in the intestine several days before it is not included in the diet. Six chickens of 40 d of age were used in the trial. The animals were fed a diet with 0.5 kg/T of ECOBIOL (*Bacillus amyloliquefaciens*) corresponding to 0.5×10^6 cfu/g before the start of the trial. At d 0 the animals were changed to a standard diet without the probiotic. Feces samples were taken during the subsequent days to be analyzed for their *B. amyloliquefaciens* content. The serial dilution method was followed as below to count the concentration

of *Bacillus amyloliquefaciens*. Five grams of sample was diluted into 300 mL of sterile saline solution (0.9%) + Tween 80 (0.4%) and homogenized in a sterile grinder (resulting in a starting dilution down to 10^{-2} to the original sample). The diluted sample was homogenized by stirring at 10,000 rpm for 1 min. Then, the sample was treated at 80°C during 1 min for keeping only the sporulated forms. Further dilutions were done by pipetting 1 mL of the diluted sample into 9 mL saline solution. Cells were counted by culturing the final dilution in Petri plates with the proper culture media, Tryptic Soy Agar (Biokar). The plates were incubated during 48 h at 30°C. Only plates containing between 30 and 300 cfu were taken into account. Results were obtained by means of at least 3 independent measurement with deviation less than 20%, from the average value. According to the results in Table 1, we can conclude that *B. amyloliquefaciens* CECT 5940 is able to persist in the gut 3 d after the additive is removed from the diet.

Table 1. Counts (cfu/g) of *B. amyloliquefaciens*

	Day 0	Day 1	Day 2	Day 3	Day 4
Animal 1	1.6×10^5	2.0×10^5	8.3×10^5	$<10^2$	$<10^2$
Animal 2	2.8×10^5	2.8×10^6	5.3×10^5	$<10^2$	$<10^2$
Animal 3	4.7×10^4	3.5×10^5	3.1×10^5	$<10^2$	$<10^2$
Animal 4	1.5×10^5	1.4×10^5	2.8×10^5	$<10^2$	$<10^2$
Animal 5	1.3×10^5	1.0×10^6	6.4×10^5	$<10^2$	$<10^2$
Animal 6	1.6×10^5	3.1×10^5	5.9×10^4	$<10^2$	$<10^2$
Mean ($\log_{10}$)	5.1^b	5.7^a	5.5^{ab}	0.0^c	0.0^c

[a–c]Different superscript letters in the same row indicate statistical differences ($P < 0.0001$).

Key Words: bacillus, probiotic, direct-fed microbial, intestine, persistence, amyloliquefaciens

110 Evaluation of a novel probiotic formulation designed to reduce decreased body weight gain in chicks following vaccination with commercially available coccidiosis vaccines.

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Coccidiosis, caused by several protozoal pathogens belonging to the *Eimeria* genus, detrimentally affects the worldwide poultry industry through increased mortality, reduced feed efficiency, reduced body weight gain, and increased susceptibility to other enteric pathogens. In an effort to reduce coccidiosis in chickens, coccidiosis vaccines may be used to protect against this disease. While effective at establishing immunity against *Eimeria* spp., the vaccine has been known to decrease early growth rate and leave chicks more susceptible to secondary bacterial infections within the gut. One potential way to ameliorate these effects may be to administer probiotic bacteria to vaccinated flocks. A hatchery applied, lactic acid bacteria based probiotic, FloraStartC (FSC), which was selected through an in vitro and in vivo screening process, was used on these trials. FSC was applied to chicks to determine the effects on body weight gain when administered concurrently with a commercially available coccidiosis vaccine. Multiple in vivo experiments were performed and, in each experiment, the birds were weighed on day-of-hatch and d 7, 10, and 14 to determine weight gain compared with non-probiotic treated chicks. In all experiments probiotic treated groups consistently had greater weight gain than non-probiotic treated groups at all time periods. Weight gain in FSC treated groups was statistically higher on each day with *P*-values for all sample days <0.05. These results were repeated using multiple commercially available coccidiosis vaccines. These results indicate that administration of the probiotic FSC leads to increased body weight gain in chicks vaccinated against coccidiosis.

Key Words: probiotic, body weight gain, coccidiosis, broiler, coccidiosis vaccine

111 Evaluation of probiotic and prebiotic effects on dairy cattle performance.

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A field trial was conducted to evaluate the effect of sequentially adding a probiotic and prebiotic to the feed ration of dairy cattle on Dairy Herd Improvement (DHI) performance measures. A probiotic, Dairyman's Choice, was added to the total mixed ration to allow an average daily intake of 100 g over a 2-year DHI test period. Then, a prebiotic, Celmanax, was also added to the total mixed ration to allow an average daily intake of 100 g over a 1-year DHI test period. Production data from the DHI records from 2 years before the test period was used as the control. Average days in milk were similar for the probiotic and control feeding periods. The probiotic did not increase actual and adjusted milk yields ($P > 0.05$), but did significantly improve DHI performance measures ($P < 0.05$) and significantly reduced antibiotic usage ($P < 0.001$). Milk fat content and protein yield was also unchanged in the presence of the probiotic ($P > 0.05$). Addition of the prebiotic significantly increased the average days in milk compared with the control feeding period ($P < 0.05$) and also increased actual milk yields ($P < 0.05$), adjusted milk yields ($P < 0.05$), milk quality ($P < 0.05$) and DHI performance measures ($P < 0.05$). These data indicate that there are benefits to using both a probiotic and prebiotic in total mixed rations of dairy cows in commercial herds to improve not only milk yield, but also other parameters that diminish production costs.

Key Words: prebiotic, probiotic, DHI, performance

Session III: Beneficial Microbes and Gut Health (continued)

200 Breaking down barriers: The impact of production stressors on gut immune and epithelial defense in the pig.

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The gastrointestinal tract is lined by a single layer of epithelial cells that play divergent roles in gastrointestinal health in animals and humans. On one hand, the epithelium must facilitate the digestion and absorption of vast amounts of nutrients and water on a continual basis to maintain body condition and promote efficient growth. On the other hand, the epithelium must serve as a barrier that selectively prevents potentially harmful luminal contents, containing bacterial, viruses, antigens, and toxins from gaining access to the mucosal immune system and systemic organs. Thus, the integrity of the intestinal barrier is critical to both animal performance and disease resistance. Stress is an inherent aspect of animal production and is a major contributor to disruption of intestinal function, impaired performance, and disease susceptibility. Moreover, stressors occurring early in life (e.g., early weaning, neonatal infections) can have long-lasting effects on intestinal function and disease susceptibility. The pathophysiologic mechanisms by which stressors impair intestinal defense barriers remain poorly understood. In a model of early weaning stress (weaning at 16–18 d of age) in pigs, we demonstrated that early weaning induces a marked intestinal stress response and increased intestinal permeability that was mediated by activation of the intestinal corticotropin releasing factor (CRF) receptor system and mucosal mast cells (Moeser et al., 2006; Smith et al., 2011; Overman et al., 2012). In addition to the acute, deleterious changes in intestinal permeability observed in the post-weaning period, we recently demonstrated lasting effects of early weaning stress as intestinal permeability disturbances were shown to persist into adulthood. Parallel studies demonstrated that early-weaned pigs display heightened sensitivity to later life production stressors and disease challenges (McLamb et al., 2012). Current investigations by our lab our focused on understanding the basic cellular interactions, focused on mast cell biology, in the stressed intestine that mediate impaired intestinal barrier function and intestinal disease resistance.

201 A late finishing *Lawsonia intracellularis* challenge is moderated by Lincomix feed medication.

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A successful *Lawsonia intracellularis* (LI) mucosal homogenate challenge model has not been previously reported in pigs greater than 7 wk of age. A total of 200 healthy 17-wk-old (72.8 ± 5.6 kg) pigs were randomly allotted to 2 treatments (10 pigs/pen) for 54 d. On d 0, all pigs were individually dosed with 3.6×10^9 ($9.0 \times 10^7 \times 40$ mL) live LI via oral gavage. Treatment 1 (Linco) received 110 ppm lincomycin in feed for 21 d, starting on d 0, followed by 3 weeks of unmedicated feed, 1 week of 44 ppm lincomycin followed by unmedicated feed until slaughter. Treatment 2 (Control) consumed unmedicated feed throughout. Fecal and blood samples were collected from 60 subset pigs (30/trt) on d 0 and 21 to confirm the presence of LI by PCR, and exposure to LI by immunoperoxidase monolayer assay (IPMA) serology. PCR analysis showed that 6.7% of fecal samples from Control and 0.0% of fecal samples from Linco pigs were positive for LI on d 0. By d 21, 93.0% of Control and 100.0% of Linco samples were positive for LI. All pigs were IPMA negative for LI on d 0. By d 21, IPMA analysis showed high (93.0%) rates of seroconversion to LI for both Control and Linco pigs, indicating a successful challenge. From d 0 to 21, Linco pigs had fewer ($P < 0.01$) days with a mild diarrhea score of 2 than Control (1.6 vs. 10.1% respectively) and more ($P < 0.01$) days with a normal diarrhea score of 1 (98.0 vs. 89.1% respectively). From d −1 to 21 Linco pigs had a greater ($P = 0.01$) ADG than Control (0.82 vs. 0.69 kg respectively). Overall ADG was numerically greater for Linco pigs than Control (0.75 vs.0.71 kg respectively). There was a trend ($P = 0.10$) toward a better overall feed conversion (G:F) for Linco pigs (0.31 vs. 0.32; Control and Linco respectively). Results showed that the LI challenge model used in this study successfully induced a late finishing ileitis break as measured by fecal PCR and serum IPMA. Furthermore, results showed a significant improvement in ADG during the first 21 d post challenge for pigs consuming Lincomix even though clinical signs (scours) were not severe. Lincomix is a registered trademark of Zoetis Inc.

Key Words: Lincomix, *Lawsonia* challenge

202 Evaluation of prebiotics and probiotics to reduce toxicity of pure and mixed-feed mycotoxins in vitro and to prevent carry-over of aflatoxin B1 in dairy cows.

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Mycotoxin mixtures in feed rations cause a wide variety of sub-lethal effects on dairy production with the most common cattle symptoms reduced feed intake, reduced milk production, reduced reproductive efficiency and a higher prevalence of disease caused by immunosuppression. Preventing mycotoxin interactions with the gastrointestinal mucosa using prebiotics and probiotics may provide an inexpensive method for decontamination of feeds. The present study aims to evaluate the mycotoxin-binding activities of commercial probiotics (BioSaf, Dairyman's Choice, MaxiPlex) and prebiotics (Celmanax, Safmannan, Bio-mos, Integral), in preventing mycotoxin-induced cell cytotoxicity using a bovine colonic cell line in a Lawn assay, in vitro. There was a significant dose-dependent reduction in aflatoxin B1, fumonisin B1 and T-2-toxin induced cytotoxicity in a lawn assay in the presence of Celmanax and Safmannan, but not in the presence of Biosaf, Dairyman's Choice, MaxiPlex, Bio-mos and Integral ($P = 0.001$). More significantly, only Celmanax consistently was 100% effective in preventing cytotoxicity associated with the mycotoxin mixtures obtained from feed rations of commercial dairy production sites experiencing mycotoxicoses. To validate this result, we examined the ability of Celmanax (50g/head/day) to stop the transfer of aflatoxin B1 from the digesta to the milk in commercial dairy cattle consuming naturally contaminated feed containing about 10 ppb aflatoxin B1. Two hundred eighty cattle from 2 production sites were sampled before receiving the prebiotic and monitored over a 90-d period. Inclusion of the

Celmanax, blocked 100% of the transfer of aflatoxin B1 from the digesta and its ultimate conversion to aflatoxin M1 to milk in 3 to 7 d ($P = 0.001$). Removal of the prebiotic resulted in the carry-over of aflatoxin B1 to aflatoxin M1 in the milk. Overall, it can be concluded that prebiotics comprised of selected yeast cell walls, such as Celmanax, can be effectively used to manage mycotoxin exposure in cattle.

Key Words: aflatoxin, fumonisin, T2-toxin, prebiotic, probiotic, milk

203 Effects of *Lactobacillus fermentum* I5007 supplementation on lipogenesis and adipose tissue distribution.

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Accumulated experiments suggest that intake of probiotic bacteria may improve the growth performance and reduce the visceral and subcutaneous fat mass. In the present study, *Lactobacillus fermentum* I5007 (*L. fermentum* I5007) was tested for its effect on improving the meat quality and exploring the undergoing mechanism as an additive in animal nutrition. Thirty-six weaned mice were randomly assigned to one of 3 treatments (n = 12). The experimental group was fed with diet containing *L. fermentum* I5007 (10^{10} cfu/kg) for 28 d. Weaned mice were fed with either basal diet or basal diet containing betaine (400 mg/kg), as the negative control or positive control. The results showed that *L. fermentum* I5007 had no effect on growth performance ($P > 0.05$) but it did have a hypoglycemic effect ($P < 0.05$). Histological results indicated that visceral adipocyte size enlargement was inhibited by both *L. fermentum* I5007 and betaine, whereas the intramuscular fat was increased in mice dorsal muscles. Consistently, *L. fermentum* I5007 also enhanced the expression of hormone-sensitive lipase (HSL) and fatty acid synthetase (FAS) in dorsal muscles ($P < 0.05$), whereas the expression of HSL and FAS was decreased in visceral and subcutaneous fat ($P > 0.05$). Furthermore, the experiment in vitro indicated that the metabolites of *L. fermentum* I5007 could inhibit the process of lipogenesis of 3T3-L1 cells, along with downregulation of adipogenic master genes. These experiments demonstrated that diet containing *L. fermentum* I5007 could regulate fat distribution by improving the fat deposition in skeletal muscle and inhibit the increase of visceral and subcutaneous fat. The finding indicates *L. fermentum* I5007 as an effective additive in improving the quality of meat.

Key Words: *Lactobacillus fermentum* I5007, probiotic, lipogenesis, adipogenic genes, adipose tissue distribution

204 Evaluation of the efficacy of *Enterococcus faecium* CECT4515 and *Bacillus licheniformis* to improve piglet performance.

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The objective of the trial was to evaluate the efficacy of Fecinor, a probiotic containing *Enterococcus faecium* CECT4515 and Proporc, a probiotic containing *Bacillus licheniformis*, on growth performance in weaned piglets. A total of 288 animals were randomly distributed, with weight and sex as a blocking factors in 48 pens of 6 piglets each, to 4 treatments (12 replicates). T1: Control; T2: (Control + 1 kg/t of feed of Fecinor); T3 (Control + 0.5 kg/t of Proporc, low dose); T4 (Control + 1 kg/t of Proporc, normal dose). No medication or Zn oxide was used in the feeds. A pre-starter diet was used from weaning (d 0) to d 14 and starter diet from d 15 to d 42. Two identical rooms were needed to house all enrolled piglets. Standard management and husbandry practices were used throughout the study. Body weight, feed intake, culling and mortality were measured. Average daily feed intake (ADFI), average daily growth (ADG), and feed conversion rate (FCR) were calculated accordingly. Furthermore, 144 pigs housed in 24 pens on the Room 1 (6 replicates per group) were fed diets mixed with chromic oxide from d 7 to 14 and from d 35 to 42. Then, fecal samples were collected daily for 3 d from every pen, to determine the apparent total-tract digestibility (ATTD) of dry matter (DM), crude protein (CP) and gross energy (GE). The statistical analyses were performed using SAS. The sex, the room, the treatment group and their interactions were included in the models as factors. Results were expressed as least squares mean (LSM) and standard error (SE). The experimental unit was the pen. Significance level was 0.05. Overall ADG of the animals fed with Proporc at low and normal doses and Fecinor were similar ($P > 0.05$) and higher than the control group (T1). ADFI was similar between groups, therefore FCR was improved by Proporc at low dose and Fecinor ($P < 0.05$) and tended to be improved with Proporc at normal dose ($P < 0.01$). ATTD of the DM, CP, and GE tended to improve with Proporc at low dose and Fecinor ($P < 0.10$) from d 40 to 42 post-weaning. No differences were detected between Proporc at normal dose and the negative control group ($P > 0.05$).

Key Words: bacillus, enterococcus, probiotic, intestine, performance, digestibility

205 Comprehensive study of swine and poultry probiotics showed high variation in quality and mode of action.

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Probiotics are today commonly used feed additives to improve the well-being and production of farm animals. Probiotics are defined as "live micro-organisms which, when administered in adequate amounts, confer a health benefit on the host." When dealing with live microorganisms the production process is crucial to ensure their survival and thus the optimal concentration, purity and stability of the final product. The identification of bacterial species is important as species differ in stability, optimal growth conditions, safety and probiotic properties. The mode of action (MOA) of probiotics is complex with competitive exclusion and pathogen inhibition as some of the major effect. The objective of this study was to evaluate selected probiotics for poultry and swine in respect to purity, concentration, identification as well as MOA such as pathogen inhibition. Most of the products were based on *Bacillus* species. A total of 14 probiotic products collected from the field were evaluated. Concentrations of the products were analyzed by serial dilution, spread plating, and counting (cfu). The purity was estimated by visual observation of morphology of non-heat treated and heat-treated products. Identification analyses were performed by isolation of colonies, DNA fingerprinting by

pulsed-field gel electrophoresis (PFGE) and sequencing and analysis of 16S rDNA. Pathogen inhibition was evaluated against 11 pathogens relevant for the swine and poultry industry by the streak method. Preliminary results show high variation in quality, purity, and efficacy between the probiotics (Table 1). It can be concluded that only a few products possesses both high quality and purity as well as relevant pathogen inhibition.

Table 1. Evaluation of probiotic feed additives A to O for poultry and swine

	Purity[1]	Concentration above label guarantee	ID according to label	Pathogen inhib.
A	Pure	Yes	OK	–
B	Pure	Yes	OK	+++
C	Pure	Yes	OK	++
D	Pure	yes	OK	+
E	Contam.	Yes	Not OK	+
F	Pure	Yes	Not OK	++
G	Contam.	Yes	Not OK	+
H	Contam.	Yes	Not OK	+
I	Pure	No	OK	+
K	Not concl.	No	NA	+
L	NA	No (no growth)	NA	NA
M	Not concl.	No	Not OK	++
N	Pure	Yes	OK	+++
O	Contam.	Yes	OK	+

[1]Contam. = contaminated; Concl. = conclusive.

Key Words: probiotic, quality, purity, pathogen inhibition

206 Applications of a probiotic and prebiotic alleviate polymicrobial mastitis in dairy cattle.

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Probiotic and prebiotic applications have recently been shown to mitigate gastrointestinal infections with Shiga toxin producing - *Escherichia coli* in all age classes of cattle. It is unclear whether similar applications can alleviate infections in peripheral tissues such as the mammary gland. Antibiotics are used for treatment of infectious mastitis in dairy cattle, but outcomes are variable due to the polymicrobial nature of the infections and greater antibiotic resistance. In this study, 2 production sites were chosen that exhibited polymicrobial mastitis that did not respond to traditional antibiotic treatments. A total of about 560 cattle were monitored before and after an application of 50 g per head per day of a prebiotic, Celmanax, and 50 g per head per day of a probiotic, Dairyman's Choice, in the feed ration. Milk samples were collected before the application began and the infections were followed over a 90 d period. Bacterial counts and diversity in the milk samples varied between the production sites. The following bacterial pathogens were present: production site A: *Aeromonas hydrophila*, *Serratia liquifaciens*, *Pseudomonas koreenis*, *Pantoea agglomerans*, Shiga toxin-producing *Escherichia coli* (STEC), *Enterobacter amnigenes*, *Staphylococcus aureus*, and *Streptococcus scirui*; production site B: *S. aureus*, *S. uberis*, *Enterococcus faecium*, *E. faecalis*, and *Pseudomonas pseudocaligenes*. All isolates grew on ESBL plates that detect extended spectrum β-lactamase-producing organisms. By d 14 after the start of the application, the prebiotic/probiotic significantly eliminated the STEC infections in the polymicrobial mastitis cases ($P = 0.01$). Over the next 76 d, there was a significant reduction in the number and diversity of the bacterial pathogens found in the milk ($P = 0.001$). By d 90, the milk samples had 0 or 1 bacterial pathogen present, but at very low numbers. All clinical symptoms were absent at 2 mo. We conclude that a prebiotic and probiotic application appears to have changed the pathogen content over time of infectious mastitis in dairy cattle. More studies are required to confirm the use as an alternative to antibiotics.

Key Words: prebiotic, probiotic, mastitis

207 Applications of a probiotic and prebiotic alleviate mammary aspergillosis in dairy cattle.

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Mastitis causes significant economic losses to the dairy industry. Prebiotics and probiotics are gaining support as alternative therapies to traditional antibiotics in managing bacterial gastrointestinal infections in cattle. In this study, we examined the effect of applying a prebiotic, Celmanax, and a probiotic, Dairyman's Choice, on the outcome of mammary aspergillosis cases. The application rate was based upon earlier positive results with bacterial-based mastitis cases using 50 g per head per day prebiotic and probiotic in the feed ration. About 240 dairy cattle were sampled over 90 d with 33% of the dairy cattle presenting with *Aspergillus fumigatus*-based mastitis at the start of the test period. Several dairy cattle succumbed to aspergillosis infections before the test period confirming the presence of fungal lesions in the lungs, intestine, and mammary glands supporting the intestine as the initial infection site. Examination of the diet confirmed high levels of *Aspergillus fumigatus* present in the haylage and barley silage. After 30 d, 9% of the dairy cattle had *Aspergillus fumigatus*-based mastitis with some clinical symptoms such as swollen hocks resolved. No new cases of *Aspergillus fumigatus* mastitis were identified. After 60 d, 3% of the dairy cattle had *Aspergillus fumigatus*-based mastitis with all clinical symptoms resolved with no new cases. After 90 d, no fungal infections were evident despite the continued presence of *Aspergillus fumigatus* in the feed ration. We conclude that administering a prebiotic and probiotic in the feed ration does manage mammary aspergillosis in dairy cattle.

Key Words: *Aspergillus fumigatus*, mastitis, prebiotic, probiotic

208 The role of a selected *Bacillus subtilis* direct-fed microbial in performance, intestinal viscosity, bacterial translocation, and bone mineralization in broiler chickens fed with high-NSP diets.

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In the present study, rye-based diets, with or without DFM, were administered ad libitum to 1-d-old broiler chickens in 3 independent experiments. In Experiments 1 and 2, chickens in both groups were humanely killed at 10 d of age and liver samples were aseptically collected to determine bacterial translocation (BT). Samples were also obtained for determination of intestinal viscosity (IV) and bone parameters. Broilers fed with rye plus DFM had a significant reduction ($P < 0.05$) in BT and IV, but bone quality was increased showing a higher breaking strength and increased Ca and P content in tibia of DFM-supplemented chickens. Experiment 3 also consisted of the same non-treated control rye diet or supplemented with DFM. In this experiment, each group consisted of 8 replicates of 20 chickens (n = 160) and 2 rye-base diets: starter (0–7d) and grower (7–28d). Each week, body weight (BW); feed intake (FI); and feed conversion rate (FCR) were determined. At d 28 BT, IV, and bone parameters were determined. Consumption of *Bacillus*-DFM improved BW and FCR. A significant reduction in BT and IV was observed in the group of chickens fed the rye-base diet with *Bacillus*-DFM when compared with the control non-treated diet. Bone quality was increased showing higher breaking strength and Ca and P contents in tibia of supplemented chickens. Additionally, in 2 independent experiments including 8% distillers dried grains with solubles (DDGS) or barley in the diet (n = 480), an improvement in performance parameters and bone quality was observed when the *Bacillus*-DFM was included in the feed. The results of these studies suggest that the consumption of a selected *Bacillus*-DFM, producing a variable set of enzymes, could contribute to enhanced performance in high NSP diets through: Reducing BT, IV, improving digestibility and bone quality. In vivo performance trials with different diets are currently being evaluated.

Key Words: *Bacillus*, direct-fed microbial, enzymes, bacterial translocation, viscosity

209 Molecular survey of the microbiomes in broilers to understand probiotics-induced reduction of BCO lameness.

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Bacterial chondronecrosis with osteomyelitis (BCO) is an important cause of lameness in commercial broiler chickens. Our previous research showed that prophylactic administration of probiotics can effectively reduce BCO lameness in broilers. In this study, we sought to understand the microbial mechanisms by which probiotics reduce BCO lameness by employing 16S rRNA gene profiling of cecal and bone microbiomes. Broilers were fed control feed or the same feed containing synbiotics BacPackTM 2X. Adding BacPack 2X to the feed on d 1 through 56 delayed the age of onset and reduced the cumulative incidence of BCO on wire flooring when compared with broilers fed the control feed (24.0% vs. 40.7%, respectively; $P = 0.003$). For microbiome analysis, cecal, proximal femur, and proximal tibia samples were collected from control (n = 4) and treatment groups (n = 4) on d 1, 17 and 56. These samples were used for microbiome analysis by deep sequencing of V1-V3 regions with MiSeq. Bioinformatics analysis of 7.7 million assembled reads revealed that complex bacterial communities exist in all samples, including the cecal and bone samples from 1 d old chicks. ANOSIM analysis showed that the groupings based on age, sample type, BacPack 2X treatment, and BCO were statistically significant ($P \leq 0.005$). PCoA plots indicated that the microbiomes in all 3 different sampling sites (ceca, femur, and tibia) were influenced by BacPack 2X treatment. Interestingly for the samples collected on d 56, BacPack 2X treatment greatly reduced the level of *Staphylococcus* in the femora and tibiae. Rarefaction curves demonstrated that the α diversity in both the femora and tibiae decreased with aging, while it increased in cecal samples. We also observed that BacPack 2X treatment significantly increased the α diversity of the bone microbiomes, which could be an indication of more stable and BCO-resistant microbiomes. Understanding the microbial species associated with BCO and influenced by probiotics treatment will help us identify opportunities for modulating the pathogenesis of BCO lameness in broilers.

Key Words: broilers, BCO lameness, probiotics, bone microbiomes

Session IV: Diversity of the Microbiome/ Impact of Gut Microbial Communities

210 Development of the microbiome of chicks: Early exposure influences future microbial diversity, independent of colonization.

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While producers are heavily invested in improving the health of American food animals, consumer pressure is driving them to do so while also minimizing the use of antibiotics. Consequently, much attention has been focused on the use of bacterial vaccines and commensal bacteria to maintain gut health; however, although evidence of their benefit to the host is mounting, little is known about how these products affect development of the gut microbiome. Using the chicken model, our laboratory evaluated the effect of a live attenuated *Salmonella* vaccine and probiotic supplementation on development of the gut microbiome. At d0 animals were vaccinated with attenuated live *S. typhimurium* (Ceva Salmune) or mock vaccinated with diluent and fed a control diet with or without a lactic acid bacteria probiotic supplement (PrimaLac; 3×10^5 cfu/g feed). Development of the cecal microbiome was assessed at d 0, 1, 3, 7, 14, and 28 by 16S sequencing of cecal digesta. Principal component analysis of the sequence reads revealed that samples cluster primarily by age, with microbiome diversity increasing over time. Individual time points show microbial differences due to vaccine status, and to a lesser extent, probiotic status. The vaccine-dependent shift in diversity occurs despite the extremely low prevalence of *Salmonella* species in the cecum (4 weakly positive samples at d 14, 0 samples at d 28). Evaluation of the taxonomic groups across treatments shows a high proportion of *Enterobacteriaceae* at d 0, 1, and 3, though these taxa drop by wk 1, and constitute only 5% of total sequences by d 28. When evaluating taxonomic groups among and between treatments, shifts in the *Clostridiales* order dominate, influenced by both vaccine and probiotic treatments. Despite its transient nature and low abundance in the cecum, *Salmonella* continues to shape the makeup of the microbiome long after it disappears. This research suggests early exposure to some bacteria may steer the microbiome in directions that leave it permanently distinct from other populations.

Key Words: microbiome, probiotics, poultry, gut development, chicken

211 Transfer of heifers calves from maternity to calf pen: The impact on the population of enterobacteria.

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The aim of this research was to evaluate the population of enterobacteria in fecal samples of calves in the maternity and calf pen. Stool samples were harvest in the maternity in less than 6 h after birth and before the administration of colostrum (T0); the second one was obtained in the calf pen between 24 and 48 h of life (T1). The material was obtained wearing sterile latex glove, directly from the rectum, then transferred to universal collectors and tubes containing sterile tetrathionate enrichment broth. Then, it was transported to the laboratory under refrigeration (4°C to 8°C). The samples were inoculated on sheep blood agar 5%, MacConkey agar and *Salmonella-Shigella* (SS); and the tubes with tetrathionate were preincubated for 24 h at 37°C, before were inoculated in SS medium. All the plates were incubated at 37°C under aerobic conditions with reading and identification of colonies after 24–48 h. Microorganisms were identified according to their growth, morpho-tinctorial characteristics and biochemical tests. At time T0, there was negative isolation in 6 stool samples (6/12, 50%). The remainder (6/12, 50%) showed isolated growth (3/6) or mixed (3/6). It was possible to identify the following bacterial strains (n = 9): *Enterobacter sakazakii, Citrobacter koseri, Morganella morganii, Proteus mirabilis, Staphylococcus* spp., *Klebsiella ozaenae* and non-fermentative bacteria (n = 3). At the time T1 it was possible to detect the presence of mixed aerobic enterobacteria isolation in all fecal samples (12/12, 100%). It was detected strains in the moment T1: *Proteus mirabilis* (n = 6), *Proteus vulgaris* (n = 3), *Escherichia coli* (n = 6), *Klebsiella azaenae* (n = 1), *Klebsiella pneumoniae* (n = 4), *Enterobactergergoviae* (n = 1), *Enterobacter agglomerans* (n = 2), *Enterobacter cloacae* (n = 1), *Enterobacter Sakazaki* (n = 1), *Salmonella enteritidis* (n = 2), *Citrobacter koseri* (n = 3) and *Morganella morganii* (n = 1). The amount of strains of isolated enterobacteria was greater when heifer calves were inside the calf pens, probably due to the contact with microorganisms of the environment and of the milk.

Key Words: neonate, development, bacteria, environment

212 *Eimeria maxima* causes changes in mRNA expression of genes associated with amino acid and sugar uptake in the jejunum of infected broilers.

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Coccidiosis in chickens is caused by infection of gut epithelial cells with protozoan parasites of the genus *Eimeria*. This disease causes significant losses to the poultry industry because infected birds fail to gain weight as rapidly as healthy birds. For the present study the effect of *Eimeria* on expression of components of amino acid and sugar uptake mechanisms was determined. Ross heritage broilers were infected with 3,000 sporulated oocysts of *Eimeria maxima*, which infects the jejunum. Sections of the jejunum were taken at d 0, 3, 5, 7, 10, and 14 post infection (PI) for mRNA expression analysis. Genes examined included digestive enzyme (APN), peptide and amino acid transporters (PepT1, $ATB^{o,+}$, $b^{o,+}AT$/rBAT, B^0AT, CAT1, CAT2, EAAT3, LAT1, y^+LAT1 and y^+LAT2), and sugar transporters (GLUT1, GLUT2, GLUT5 and SGLT1). At d 7 PI when the pathology is greatest, the weight gain of infected birds was approximately 50% of the controls. Similarly, the effects on gene expression were also greatest at that time point. Gene expression of APN, PepT1, $b^{o,+}AT$/rBAT, B^0AT, and EAAT3, all of which encode proteins located on the brush border surface, were significantly decreased at d 7 PI. The expression of one gene ($ATB^{o,+}$) which encodes an amino acid transporter (AAT) on the brush border surface did not change during the course of the infection. The expression of AATs located on the basolateral surface did not show as large a decrease in expression as did the brush border AATs. In fact, CAT1 and

LAT1 showed increased expression at d 5 and 7 PI, respectively. The expression of the 4 sugar transporters was significantly decreased at d 7 PI. In conclusion, *E. maxima* infection caused a general decrease in gene expression of sugar transporter and brush border AATs at d 7 PI. By d 14 PI expression of all genes did not significantly differ from controls. It is possible that decreased expression of amino acids and sugar transporter genes leads to a decrease of nutrients coming into gut epithelial cells, which can limit nutrients available to the parasite and thus inhibit its reproductive and infective capacities.

Key Words: *Eimeria*, amino acid transporter, sugar transporter, gene expression, broilers

213 Interrelationships of fungal and bacteria populations within the gastrointestinal tract of poultry.

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Effective *Salmonella* control in broilers is important from the standpoint of both consumer protection and industry viability. We investigated associations between *Salmonella and Campylobacter* recovery from different sample types collected at sequential stages of grow-out from the broiler gastrointestinal tract and production environment. The goal of the present study was to record changes in fungi populations recovered from poultry gastrointestinal tracts and relate those changes to foodborne pathogen status. Over 3000 broiler gastrointestinal samples were isolated and over 680 samples were further characterized using an automated repetitive sequence based PCR (rep-PCR) methodology to track fungal genera changes during successive grow-outs. Over 24 different fungal and yeast genera were identified using rep-PCR including *Rhizopus* spp., *Aspergillus* spp., *Penicillium* spp., and *Fusarium* spp. The results from the present study will provide a normal fungi genera under commercial conditions, relate these fungi to foodborne pathogens, and will be a stepping stone for investigating the effect of fungi on the gastrointestinal tract and overall health of poultry.

Key Words: fungi, *Salmonella*, *Campylobacter*, poultry

214 Modification of the chicken cecal microbiome by *Campylobacter jejuni* colonization and by a feed additive.

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Campylobacter jejuni is a foodborne pathogen causing severe enteritis in humans with chickens being the most important identified source of *C. jejuni*. It colonizes the chicken cecum up to 10^9 cfu/g of cecal matter. Despite this, chickens were rarely proven affected by *C. jejuni* presence. The effect of such intense colonization on the chicken cecal microbiome is unknown. Efforts are made to control this pathogen at the farm and some in-feed control measures are showing encouraging results. Modifications of the chicken intestinal microbiome by these measures are often hypothesized as part of their mechanism of action. In this study, 4 groups of 15 chickens were used. Chickens received or not a feed additive, based on a protected mix of organic acids and essential oils, tested as a *C. jejuni* control option, from hatch to the end of the experiment. Fourteen days old chickens were then infected or not with *C. jejuni*. Birds were euthanized at 35 d of age. Cecal content from each chicken was recovered. *C. jejuni* cecal levels were determined by culture on mCCDA. DNA was also extracted to perform in-depth microbiome analysis. Levels of *C. perfringens*, *E.coli*, lactobacilli, enterobacteria, and *Bifidobacterium* were evaluated by real time PCR. In each group, DNA from 8 chickens was subjected to 16S rDNA sequencing using the Ion Torrent technology. Sequences analysis was performed with Mothur and the Greengenes database. The feed additive lowered the *C. jejuni* presence in the chicken cecum by 0.6 log. *C. jejuni* colonization was associated with increased *Bifidobacterium* levels. Alpha-diversity was not much affected by *C. jejuni* presence but β-diversity was. The relative abundance of the phylum composing the cecal microbiome of *C. jejuni* colonized chickens was different from the one composing the *C. jejuni* negative birds but this change was unexpectedly mild. The feed additive did not affect the chicken cecal microbiome diversity but lowered *Streptococcus* relative abundance. Overall, these results show that *C. jejuni* does not greatly disturb the chicken cecal microbiome and that the feed additive affected *C. jejuni* counts with no effects on the cecum diversity.

Session V: Nutrition and Gut Health

300 Impact of a multistrain *Bacillus* product on broiler performance and small intestinal microbiota.

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Direct-fed microbials (DFM) have been reported to improve nutrient digestibility and affect the gut microbial ecosystem by shifting the balance of beneficial and harmful organisms. DFM have been known to contribute to improved weight gain and/or feed efficiency in broilers. The objective of this work was to determine if the addition of a combination of a multistrain *Bacillus subtilis* product (DFM) to a feeding program improves performance and changes the gut microbiota of young and market age broilers fed corn/soy/DDGS diets. In this 42-d floor pen study using a 3-phase feed program, 0% and 0.05% inclusion of DFM was tested. Each dietary treatment was replicated 8 times using 60 male Cobb 500 broiler chicks per pen. The litter was 50% new wood shavings and 50% litter from a previous study. At 15 and 42 d of age, 2 birds per pen were randomly selected and pooled for small intestine mucosal microbiota assessment. There were no differences in 42-d gain and feed intake between treatments. Feed efficiency and caloric conversion tended to improve (P = 0.06) due to DFM addition. Small intestine mucosa of DFM birds at 15 d of age had 18% increase in *Lactobacillus* (as a percent of total lactic acid bacteria, tLAB; $P < 0.05$) compared with birds without DFM supplementation. In contrast, *Enterococcus*, as a percent of tLAB, was 18% lower ($P < 0.05$) in birds with DFM compared with birds without DFM supplementation at the same age. Despite a low *C. perfringens* count in small intestine mucosa of the control birds, there was a significant reduction observed in this parameter in the DFM birds at 42 d of age. These results indicate that feed and energy conversion of broilers was improved due to the inclusion of DFM. Additionally, gut microbiota was affected by a specific multi-strain DFM differently at 15 and 42 d of age.

Key Words: direct-fed microbial, broiler, *Lactobacillus*, *Enterococcus*, *C. perfringens*

301 Utilization of rye as energy source affects bacterial translocation, intestinal viscosity, microbiota composition and bone mineralization in broiler.

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Two independent trials were conducted to evaluate the utilization of rye as energy source on bacterial translocation, intestinal viscosity, gut integrity, gut microbiota composition, and bone mineralization, when compared with a traditional cereal (corn) in broiler chickens. In each experiment, day-of-hatch, broiler chickens were randomly assigned to either a corn or a rye diet (n = 20 chickens/group). At 10d of age, in both experiments, 12 chickens/group were randomly selected, and given an oral gavage dose of fluorescein isothiocyanate dextran (FITC-d). After 2.5 h of oral gavage, blood samples were collected to determine the passage of FITC-d. The liver was collected from each bird to evaluate bacterial translocation (BT). Duodenum, ileum and cecum gut sections were collected to evaluate intestinal viscosity and to enumerate gut microbiota. Tibias were collected for observation of bone parameters. Broilers fed with rye showed increased ($P < 0.05$) intestinal viscosity, BT, and serum FITC-d. Bacterial enumeration revealed that chickens fed with rye had increased the number of total lactic acid bacteria (LAB) in all 3 sections of the gastrointestinal tract evaluated when compared with chickens fed with corn. Chickens fed with rye also had significantly higher coliforms in duodenum and ileum, whereas the total number of anaerobes increased only in duodenum. A significant reduction in bone strength and bone mineralization was observed in chickens fed with rye when compared with corn fed chickens. In conclusion, rye evoked mucosal damage in chickens that alter the intestinal viscosity, increased leakage through the intestinal tract, and altered the microbiota composition as well as bone mineralization. Studies to evaluate dietary inclusion of selected DFM candidates that produce exogenous enzymes in rye fed chickens are currently being evaluated.

Key Words: bacterial translocation, intestinal viscosity, rye, bone mineralization, chickens

302 Impact of dietary exogenous enzyme supplementation on enteric adherent mucin thickness layer and gastrointestinal morphological development in poultry.

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Anti-nutritional factors in feed ingredients (ANF) can reduce nutrient utilization and suppresses gut health. Birds typically activate their innate immune system for protection against the adverse effects of ANF, which often involves the secretion of mucin. Although dietary supplementation of exogenous enzymes are commonly used to alleviate the adverse effects of ANF on apparent nutrient digestibility, little is known about how they affect gut health, particularly in relation to enteric mucosa morphological development, and the mucin secretion. We carried out 5 studies to examine the effect of dietary supplementation of different enzymes on gut health, by accessing the effect of jejunum morphological development and ileal enteric adherent mucin thickness layer in turkeys and broilers. Dietary supplementation of a blend of XAP enzymes (xylanase, amylase, and protease) improved apparent nutrient digestibility and reduced ileal adherent mucin layer thickness. Dietary β-mannanase supplementation also improved the jejunum villi development and lowered the ileal adherent mucin thickness layer, thus reducing the endogenous loss of nutrients. Phytase supplementation significantly reduced the ileal adherent mucin thickness layer and improved nutrient digestibility. In conclusion, dietary supplementation of exogenous enzymes helped alleviate the adverse effects of ANF on nutrient utilization by directly or indirectly removing the mucosal irritation that stimulates enteric mucin secretion.

Key Words: enzyme, gut health, morphology, mucin

303 Phytogenic feed additives as replacement for antibiotic growth promoters in broiler chickens.

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The recent trend toward reduction of antibiotic growth promoters (AGP) in North American poultry diets has put tremendous pressure on the poultry industry to look for viable alternatives. In this context, phytogenic feed additives (PFA) are researched to improve gut health and there by performance. An experiment was conducted with the objective to evaluate the effects of PFA as a replacement for AGP on the small intestinal histomorphology, cecal microflora composition, nutrient digestibility and growth performance in broiler chickens. A total of 432, day-old Cobb 400 broiler chicks were randomly assigned to replicate pens of 3 treatments with each consisting of 12 replicate pens (n = 12 chicks/pen). The chicks were fed a corn-soybean meal based control (CON), CON + 500 mg/kg of AGP (bacitracin methylene disalicylate containing 450 mg active BMD/g), and CON + 150 mg/kg of proprietary blend of PFA (Digestarom Poultry) until 39 d of age when samples were collected. Birds fed either AGP or PFA had increased villus height in all 3 segments of the small intestine in comparison to the birds fed CON ($P \leq 0.05$). Furthermore, the PFA fed birds had significantly increased villus height and lower crypt depth compared with AGP fed birds ($P \leq 0.05$). Birds fed both the additives had also increased total-tract digestibility of dry matter, crude protein and ether extract ($P \leq 0.05$). This strongest effect of the PFA on villus height in the jejunum, where majority of absorption occurs, may suggest efficient nutrient absorption in PFA fed birds. Although both the additives reduced the total cecal counts of *Salmonella* spp., anaerobic bacteria and *Clostridium* spp., PFA alone reduced the total coliform count while increasing the counts of *Lactobacillus* spp. ($P \leq 0.05$). This data suggests the establishment of beneficial microbial colonies in the PFA fed birds. Overall, both PFA and AGP increased the body weight gain while lowering the feed conversion ratio ($P \leq 0.05$). Hence, data from this experiment demonstrated the efficacy of PFA in place of the AGP in poultry diets.

Key Words: histomorphology, microflora composition, nutrient digestibility, performance

304 Gut health model using different diet formulations in broiler chickens.

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A healthy gastrointestinal (GI) tract is important for optimal production performance and reduced illness caused by translocation of enteric bacteria to various organs. Oral administration of fluorescein isothiocyanate dextran (~4 kDa; FITC-d) and measuring its translocation through tight junction leakage via serum levels is an effective indicator of gut health in poultry. In our laboratory, studies conducted in chicks showed that rye-based diet and 24h feed restriction can cause increased serum FITC-d levels, indicating increased gut leakage. Furthermore, raw soybeans contain various anti-nutritive factors (ANF) which may be retained if SBM is not processed to optimal conditions. Therefore, we recently conducted study to determine the effect of such ANF factors on production performance and enteric tight junction integrity in birds. Day-old chicks were randomly assigned to treatments (n = 15 birds/treatment) and grown for 2 weeks of age. Treatment (TRT) 1 was a control group provided diet containing FFSB autoclaved for 1h, TRT 2 was given diet with an equal combination of both autoclaved and raw FFSB, and TRT 3 received diet containing raw FFSB. Autoclaving reduced trypsin inhibitors (TI) in FFSB from 45.3 to 11.7 mg/g and resulted in TRT 1, 2, and 3 diets containing 2.4, 10.1, and 19.4 mg/g of TI, respectively. Results from the trial showed that as the level of raw FFSB increased in diet, there was lower ($P < 0.05$) body weight and gain among the treatment groups. Also, TRT 2 and 3 had increased ($P < 0.05$) pancreas weight expressed as percent of body weight, suggestive of pancreatic hypertrophy to compensate reduced protease activity in GI tract due to higher levels of trypsin inhibitors in diet. Both TRT 2 and 3 showed higher ($P < 0.05$) serum FITC-d levels compared with TRT 1 birds indicating an increase in gut tight junction leakage with raw FFSB in diet. In fact, there was a negative correlation ($r^2 = 0.56$) between body weight at 14d and serum FITC-d levels. Thus, we can conclude that presence of ANF in soybeans had a negative effect on gut integrity and reduced production performance in birds. In the future, this gut health model can be used to evaluate different dietary, nutritional, and intervention strategies.

Key Words: gut inflammation, broilers, soybean meal, FITC-d, tight junction leakage

305 Protein-mediated butyrate transport in the rumen epithelium is modulated by feed restriction in Holstein steers.

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The objective of this study was to elucidate the role of protein-mediated transport mechanisms in the transport of acetate and butyrate across the rumen epithelium during subacute ruminal acidosis and feed restriction. Individually housed Holstein steers (n = 21) were fed a total mixed ration ad libitum for 5 d, then assigned one of 3 treatments. The control group (CTRL) was fed ad libitum for 1 d. The acidosis group (ACID) received 25% of baseline DMI on d 1 and a 30% DMI barley grain challenge on d 2. The feed restriction group (FR) received 25% of baseline DMI for 5 d. Reticuloruminal pH was measured continuously for the treatment period. Using Ussing chambers, flux rates of acetate and butyrate were measured under 3 buffering conditions: bicarbonate-containing buffer, bicarbonate-free buffer with nitrate, and bicarbonate-free buffer with nitrate and pHMB, an MCT inhibitor. Total flux, protein-mediated flux (PMF), MCT-mediated flux (MCT) and passive diffusion flux (PDF) were calculated. Duration of subacute ruminal acidosis was significantly higher in ACID compared with CTRL and FR (57 ± 90 vs. 519.71 ± 90 min/d vs. 30 ± 90 min/d for CTRL, ACID and FR; $P < 0.01$). Total acetate flux was higher in FR than in CTRL (421.1 ± 41.4 vs. 630.6 ± 38.9 nM/cm^2·h, $P < 0.01$), but there

was no difference between CTRL and ACID (421.1 ± 41.4 vs. 455.4 ± 38.9 n*M*/cm^2·h). Total butyrate flux also increased in FR compared with CTRL (625.5 ± 86.3 vs. 1241.9 ± 94.8 n*M*/cm^2·h; $P < 0.01$), but was not significantly different between CTRL and ACID (625.5 ± 86.3 vs. 716.7 ± 81.0 n*M*/cm^2·h). Flux through MCT was not significantly different among CTRL, ACID and FR treatments for acetate (78.5 ± 38.9 vs. 5.8 ± 38.9 vs. 19.9 ± 41.3 n*M*/cm^2·h for CTRL, ACID and FR) or butyrate (76.1 ± 86.1 vs. 36.1 ± 81.0 vs. 14.2 ± 94.8 n*M*/cm^2·h for CTRL, ACID and FR). In butyrate, PMF was significantly higher in FR than in CTRL (99.9 ± 86.3 vs. 479.21 ± 103.9 n*M*/cm^2·h, $P < 0.01$), but there was no difference between CTRL and ACID treatments (99.9 ± 86.3 vs. 90.2 ± 81.0 n*M*/cm^2·h). These results suggest increased butyrate flux during feed restriction is partly due to a protein-mediated flux mechanism.

Key Words: rumen epithelium, butyrate transport, feed restriction, subacute ruminal acidosis

306 Effects of Sporulin and Cibenza CSM on gut health and growth performance of broilers.

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The gastrointestinal tract is constantly exposed to a wide variety of potentially harmful substances. Gut health is highly associated with growth performance and structural health of broilers. It is widely accepted that probiotics can counteract dysbiosis and improve gut health. Feed enzymes have been shown to improve gut health by reducing undigested substrates, anti-nutritional factors, and altering microbiota. The objective of this study was to determine the interaction between Cibenza CSM (a blend of endo-xylanase, β-glucanase, α-galactosidase) and Sporulin (a *Bacillus* spore-based direct-fed microbial) on gut health, growth performance, and structural health of broilers. This trial is a 2 × 2 factorial arrangement of treatments containing 2 levels of Sporulin (0 or 250 g/ton) and 2 levels of Cibenza CSM (0 or 500 g/ton). A total of 1,440 Ross 308 male broiler chicks were randomly assigned to 4 treatments with 6 pens per treatment and 60 birds per pen. All chicks were fed a corn-soybean meal based diet from hatch to d14 of age followed by rye-wheat based diets until d 56 of age. Sporulin numerically improved FCR ($P = 0.115$) and performance index ($P < 0.1$) during d 0–15, which carried over to d 29. Cibenza CSM improved ($P < 0.0001$) BWG, feed intake, FCR and performance index when birds were fed rye-wheat diets during d 15–29. Both Sporulin and Cibenza CSM reduced ($P < 0.05$) digesta viscosity, improved ($P < 0.05$) duodenum and jejunum morphometry. Sporulin reduced ($P < 0.05$) the incidence of severity of tibial head necrosis and tibial dyschondroplasia. Cibenza CSM reduced d-42 litter moisture ($P < 0.01$), d-28 serum endotoxin levels in the absence of Sporulin ($P < 0.05$) and α-1-acid glycoprotein levels in the presence of Sporulin ($P < 0.05$). In summary, both Cibenza CSM and Sporulin improved gut health, which was associated with better growth performance. Sporulin is a trademark of Pacific Vet Group-USA Inc. Cibenza is trademark of Novus International, Inc.

Key Words: Cibenza CSM, Sporulin, FCR, gut health, digesta viscosity, gut morphometry

Poster Presentations

P100 The effectiveness of direct-fed microbial and prebiotic on histomorphology of intestine, ultra-structural changes of intestinal mucosa and performance of turkey poults infected with *Salmonella* and *Campylobacter*.

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Salmonella and *Campylobacter* are considered as major public health burdens worldwide. Poultry are known to be one of the main reservoirs for these zoonotic pathogens. This study was conducted to evaluate the effect of Calsporin a probiotic or direct-fed microbial (DFM) and IMW50 a prebiotic mannan oligosaccharides (MOS) on performance, reducing *Salmonella* and *Campylobacter* colonization in the digestive tract, intestine histomorphology, and ultrastructural changes of intestinal mucosa in turkey poults. A 21-d battery cage study was conducted using 4 dietary treatments including: a basal diet as negative control (NC); basal diet supplemented with 0.05% DFM; basal diet supplemented with 0.05% MOS; and basal diet supplemented with 0.05% mixture of DFM and MOS. Three hundred thirty 6-d-old female Large White Turkey poults were randomly distributed in 6 electric heated battery cages with 12 treatments of 4 replicates per treatment containing 7 poults per pen. The first 16 pens were not infected with bacteria, poults in pens 17–32 were orally challenged at d 7 with 1 mL of 10^5 cfu/mL *Salmonella* Heidelberg, and the poults in pens 33–48 were orally challenged at d 7 with 1 mL of 10^5 cfu/mL *Campylobacter jejuni*. Feed intakes, body weight gain and FCR were measured weekly and at the end of the experiment. At d 21, fresh fecal samples from each pen were collected for *Salmonella* Heidelberg and *Campylobacter jejuni* enumeration, and ileal tissue samples were collected from one bird per pen for histomorphometry and ultrastructural examination. Results showed significant ($P \leq 0.05$) increase of BW, reduction of *Salmonella* Heidelberg in fecal samples, increased surface area of villi, increased length of villi, increased number of goblet cells, more mucus secretion, and presence high number of segmented filamentous bacteria in DFM and MOS supplemented groups compared with control groups. The results suggest Calsporin and IMW50 enhance ilealmucosal health and performance of turkey poults.

Key Words: direct-fed microbial, mannan oligosaccharides, *Salmonella*, *Campylobacter*, turkey poults

P101 A review: Alternatives to antibiotic use for growth promotion in poultry.

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Antibiotics have been used as growth promotion (sub-therapeutic doses), disease prevention (prophylactic doses) and for the treatment of infection in livestock and poultry for over 50 years. Years of research and practical experience have shown that antibiotic use significantly improves animal performance and health status. However, the use and misuse of antibiotics in feed have led to problems with drug residues in animal products and increased bacterial resistance in humans. As a result, the sub-therapeutic use of antibiotics has been banned in European countries since January 2006 and other countries are seriously considering a similar ban. Therefore, alternatives to antibiotics are urgently needed. The aim of this paper is to review the advantages and disadvantages of a wide range of products that have been tested as potential alternatives to antibiotics in farm animals (swine, poultry and ruminants) such as; organic acids, prebiotics, probiotics, herbal extracts, enzymes, bacteriophages and antibodies.

Key Words: antibiotics, probiotics, prebiotics, organic acids, herbal extracts

P102 Selective isolation of gut lactic acid bacteria from commercial beef cattle originating from farms at risk of subacute ruminal acidosis (SARA).

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The aim of this study was to identify possible probiotic lactic acid bacteria (LAB) from healthy beef cattle on farms with high concentrate feeding at high risk of SARA. Six finishing beef farms situated in Aberdeenshire were selected as being at high (n=3) or low risk (n=3) for SARA incidence, based on the starch content of the diet. Ninety-eight continental crossbreed steers and heifers of an average deadweight of 374 kg were assessed at slaughter for rumen wall damage, and samples were taken of ruminal digesta (RC), cecal contents (CC), rumen wall (RW) and caecum wall (CW). *E. coli* viable counts were made with RC and CC. Thirty-eight cattle were further selected based on the (1) high risk of SARA status of the farm, (2) low rumen damage scores, and (3) low SARA *E. coli* counts. Samples from RW (n = 38), CW (n = 20), RC (n = 2) and CC (n = 9) were pre-enriched in MRS broth supplemented with 0.3% bile. Ten-fold serial dilutions were inoculated onto MRS and modified MRS with bromophenol blue (mMRS-BPB) agar. The suitability of the mMRS-BPB media for LAB differentiation was assessed against several strains of 10 LAB species, including *Lactobacillus salivarius, L. ruminis*, *L. amylovorus*, *L. reuteri*, *L. buchneri* and *L. delbrueckii*. A double layer inhibition test with a pool (1.5×10^8 cfu/mL) of 4 SARA *E.coli* strains was applied to both MRS and mMRS-BPB agar plates showing growth of 10 to 30 colonies of LAB. In total, 656 colonies displaying clear inhibition zones and distinct colony morphologies on mMRS-BPB agar were predominantly isolated from RW (n = 408) and CW (n = 181). Partial 16S rRNA gene sequencing of 508 LAB strains revealed *Lactobacillus mucosae* as the predominant (87.2%) LAB species in all types of samples, with others including *E. faecium* (4.5%), *L. salivarius* (1.77%), *P. acidilactici* (1.37%), *L. coryniformis* and *L. sakei* (0.98%), and *L. amylovorus* and *L. curvatus* (0.78%). This new assay allows

direct screening for, and selective isolation of, bile resistant LAB strains with inhibitory activity against SARA intestinal *E.coli.*

Key Words: subacute ruminal acidosis, beef cattle, probiotic, lactic acid bacteria, assay

P103 Dynamics of the microbiome over the rearing period in two lines of broilers.

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Probiotics are typically delivered to broilers at hatch to provide protection from pathogen colonization and hasten maturation. However, delivery at hatch may only provide a temporal effect because populations in the gut change due to factors such as changes in diets. Thus, probiotic cultures can be outcompeted and lost early in the rearing period. Understanding the population dynamics over a rearing period can give information regarding microbial populations that can be used for intelligent design of treatments throughout the rearing period to improve gut health. To determine microbiome dynamics, experiments were conducted where fecal samples from 2 lines of broilers were analyzed using Illumina sequencing to identify the bacterial populations. Samples were obtained 6 times throughout the rearing period. Line (A) was sampled at 0, 2, 4, 5, 6 and 16 wk of age, whereas Line B was sampled at 0, 2, 4, 7, 8 and 18 weeks of age. The 16SrRNA gene sequences were processed with Qiime and statistical analyses were conducted in R. The data from each line were analyzed separately. At the taxonomic levels of phylum, class, and order, similar trends in dynamics were seen in the 2 lines at each sampling period; aerobic bacteria (Proteobacteria) dominated at hatch and were subsequently replaced by anerobes (Firmicutes) which endured for the remaining sampling times. However, at the family and genus levels, differences between the 2 lines were apparent. At 2 wk of age, Lactobacillaceae was prevalent in both lines, but line B possessed sub-populations of Dehalobacteriaceae (range 3.5 to 18.8%) that were absent from line A. In Line A, populations of Ruminococcaceae appeared in all birds at 6 wk of age at an abundance of >15% and were similar at 16 wk of age. This family appeared in only 2 birds of Line B, at 4 wk of age, and at a lower prevalence. Small sub-populations of bacteria were also line specific. The data indicate microbial populations in broilers are dynamic and design of gut health treatments should take into account these changes and that microbes may be specific to different lines of birds.

Key Words: dynamics, age, broiler, microbiome

P104 Neonatal lambs gastrointestinal tracts are initially colonized by a unique and dynamic vaginal microbiota but rapidly transition toward the dam's teat.

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Given the well-described roles of human vaginal microbiota in reproductive outcomes and neonatal health, we have begun characterizing the vaginal microbiota of cattle and sheep and examining their contributions to neonatal health and microbiological development of the gastrointestinal tract. Culture-independent 16S rRNA gene surveys have revealed both bovine and ovine vaginal microbiota to be unique from previously described vaginal microbiota. Each has a low abundance of vaginal lactobacilli species (both <1%) and have high abundances of *Aggregatibacter* spp. (21 ± 6% and 29 ± 7%, respectively), and *Streptobacillus* spp. (13 ± 6% and 23 ± 8%, respectively). The majority of the cow and ewe vaginal samples also contained archaea predominantly from the Desulfurococcales order. Temporal analyses of ewe vaginal microbiota from breeding to parturition, revealed the ewe vaginal microbiota exhibits correlative changes in microbiota with the observation of vaginal mucous (ANOSIM $r = 0.16$, $P \leq 0.05$). Fifteen microbial taxa exhibited positive rank and/or linear relationships with progesterone concentrations. These included *Brevundimonas* spp. (Spearman's $r = 0.54$, $P < 0.05$) and *Lactobacillus spp.* (Pearson's $r = 0.33$, $P < 0.05$). Progesterone concentrations also had a significant negative correlation with vaginal pH (Pearson's $r = -0.39$, $P < 0.05$), that was most strongly linked to *Lactobacillus* spp. (Pearson's $r = -0.30$, $P < 0.05$), and *Stentrophomonas* spp. ($r = -0.33$, $P < 0.05$). Immediate post-parturition samples of ewe-lambs revealed that the dam's vaginal microbiota are the primary inoculants of the oral, nasal, and rectal orifices of newborn lambs. Following exposure to the dam, the lambs' oral, nasal, and rectal microbiota rapidly transitions from a predominantly vaginally derived microbiota to a community more similar to the dam's teat surface.

Key Words: cattle, sheep, vaginal microbiota, neonate

P105 Variations of the microbiome among sheep breeds on two different diets.

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Current research on the gut microbiome has focused on identifying signature populations associated with specific phenotypes in the ruminant animal for traits such as feed efficiency, methane production, and growth rate. The effect of breed on the microbiome has been assessed in only a few studies. The objective of this study was to evaluate the contributions of diet and breed to variation of specific taxa in the microbiome. Six Rambouillet, 6 Hampshire, and 4 Suffolk wethers were fed either a concentrate- or forage-based diet for 49 d post weaning. Rumen fluid samples were collected at the end of the feeding period. DNA extracted from the rumen fluid samples was sequenced on Illumina's HiSeq 2000 platform. Filtered reads were compared with a 16S rDNA gene database, and reads that aligned with a ≥ 97% sequence identity were clustered into operational taxonomic units (OTU). OTU abundance was fitted to a general linear model including the effects of breed, diet, and breed-by-diet interaction to determine the proportion of variation accounted for by each variable in the model. A covariate of total number of reads for each wether was also fitted to account for differences in efficiency of sequencing. A total of 142 OTU were identified, and an average of 15%, 11%, and 10% of the variation in model was explained

by diet, breed, and breed-by-diet interaction, respectively. A large amount of the variation was explained in some of the OTU by one variable of the model. For example, diet accounted for 70% of the variation in *Butyrivibrio fibrisolvens*, Breed accounted for 64% of the variation in *Anaerorhabdus furcosa*, and breed-by-diet interaction accounted for 44% of the variation in *Mitsuokella jalaludinii*. Diet accounted for the largest part of the variation in 59 of the OTU. Breed accounted for more variation than the other variables in the model in 47 OTU, and 36 OTU's variation was explained by the breed-by-diet interaction. As expected, this study shows that diet accounted for the largest variation in the sheep microbiome; however, breed and breed-by-diet interactions affect the microbe populations in certain OTU.

Key Words: diet, breed, sheep, microbiome

P106 A calf-rearing model for exploring the influence of colostrum on the microbiological health of the developing bovine intestinal tract.

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Enteric disease is a major cause of morbidity and mortality in young calves. In recent years, studies have revealed that beneficial commensal microbes contribute to intestinal health by supporting epithelial function, host metabolism, and immune development. Microbial colonization begins with the acquisition of pioneer organisms from the dam. After this, composition of the microbiome varies but the most desirable trajectory is toward increased richness and diversity. In this study, we present a calf rearing system model developed to help study the factors influencing the bovine neonatal microbiome during the pre-weaning period. The model was designed specifically to help investigators examine the role of colostrum in choreography of the neonatal calf microbiome. A total of 12 healthy male Holstein calves were separated from the cow immediately following birth, and fed 4 L of aseptically collected, high quality colostrum. All calves were housed, monitored, and fed separately for the remainder of the experiment. A group of 3 animals were euthanized for necropsy, and intestinal samples collected before colostrum administration (d 0) and progressively during the rest of the trial (d 3, 7 and 21). Intestinal mucosal and luminal samples were collected from the duodenum, jejunum, ileum and colon. The microbiome of these samples were analyzed using V3-V4 16S rRNA gene sequencing via MiSeq (Illumina) sequencing technology. Colostrum samples were distinct from duodenal, jejunal, and ileal samples though *Abiotrophia* spp. were common to all samples. Duodenal samples were near-exclusively colonized by *Abiotrophia* spp., but greater diversity was seen in the distal regions of the small intestine. Gradual changes in gut microbiota were seen with age and indicated a developing microbial ecosystem consistent with previous reports.

Key Words: calf, colostrum, microbiome

P107 Genetic and genome analyses of bacteria cultured from lame broilers with osteomyelitis.

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Lameness is a significant problem resulting in millions of dollars in lost revenue annually. In commercial broilers, the most common cause of lameness is bacterial chondronecrosis with osteomyelitis (BCO). We are using a wire flooring model to induce lameness attributable to BCO. We used 16S ribosomal DNA sequencing to determine that *Staphylococcu*s spp. were the main species associated with BCO. *Staphylococcus agnetis*, which previously had not been isolated from poultry, was the principal species isolated from the majority of the bone lesion samples. *Staphylococcus* spp. were also isolated from the blood of apparently healthy broilers. Administering *S. agnetis* in the drinking water to broilers reared on wire flooring increased the incidence of BCO 3-fold when compared with broilers drinking tap water (P = 0.001), conclusively demonstrating bacterial translocation across the gut. We found that the minimum effective dose of *Staphylococcus agnetis* to induce BCO in broilers grown on wire flooring experiment is 10^5 cfu/mL. More severe lesions were found in the proximal femoral and tibial heads in the lame birds that received *S. agnetis* in the water compared with the control group. We sequenced and assembled a draft of the *S. agnetis* genome for further investigation of genetic diversity, toxins, and pathogenicity determinants, for this poorly characterized species. Isolating pathogenic bacterial species, defining their likely route of transmission to broilers, and genomic analyses will contribute substantially to the development of measures for mitigating BCO losses in poultry.

Key Words: intestinal translocation, lameness, *Staphylococcus*, broiler, bacteremia

P108 Proteins involved in intracellular pH regulation in rumen epithelial cells are modulated during the transition period in Holstein dairy cows.

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This study examined changes in SCFA transport protein abundance in the rumen epithelium during the transition period in dairy cows. Twelve cannulated primiparous and multiparous cows were fed a total mixed ration with 46% NDF and 34% NFC during the dry period and a total mixed ration with 34% NDF and 43% NFC during early lactation. Rumen biopsies were taken 3 weeks before estimated calving (PRE), 1 week after actual calving (PERI) and 6 weeks after actual calving (POST), and were analyzed via immunofluorescence for abundance of monocarboxylate cotransporter, isoform 1 (MCT1), sodium/bicarbonate cotransporter, isoform 1 (NBC1), sodium/proton exchanger, isoform 3 (NHE3) and carbonic anhydrase, isoform 2 (CA2). Immunofluorescence analysis showed no significant differences among the PRE, PERI and POST time periods for MCT1 (16878 ± 1555 AU, 15613 ± 1508 AU, and 16886 ± 1555 AU, respectively), NBC1 (16467 ± 1274 AU, 13983 ± 1318 AU, and 15047 ± 1324 AU, respectively) and NHE3 (10467 ± 1066 AU, 9465 ± 1066 AU, 8552 ± 1109 AU, respectively). Abundance of CA2 was downregulated from PRE to PERI and POST (12332 ± 1580 AU vs. 7235 ± 1580 vs. 8121 ± 1643, respectively; P = 0.01, PRE vs. PERI). These results suggest that increases in epithelial surface area, coupled with no significant changes in MCT1, increase the epithelial SCFA transport capacity during the parturition transition. Further, the lower abundance of CA2 following parturition may contribute to intracellular acidification

of epithelial cells to promote epithelial remodeling during early lactation.

Key Words: rumen epithelium, transition period, transport protein, intracellular pH

P109 High and low loads of cecal colonization by *Salmonella* Enteritidis in chickens triggers distinct immune kinome profiles.

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Salmonella enterica serovar Enteritidis are facultative intracellular bacteria that cause disease in numerous species. *Salmonella*-related infections originating from poultry and/or poultry products are a major cause of human foodborne illness, and *Salmonella* Enteritidis is the leading cause worldwide. Despite the importance of *Salmonella* to human health and chickens being a reservoir, little is known of the response to infection within the chicken gastrointestinal tract. Using chicken-specific kinome immune peptide arrays we compared a detailed kinomic analysis of the chicken gut immune response in birds with low and high *Salmonella* loads. Four-d-old chickens were challenged with *S.* Enteritidis (10^5 cfu) and cecal content and a section of jejunum collected on d 4, 7, 10, 17, 24 and 37 post-infection (pi). *Salmonella* colonization was enumerated and birds with the lowest and highest loads at each time point were selected for kinomic analyses. The most dramatic changes between birds colonized with low and high loads of *Salmonella* Enteritidis were observed within the first 17 d pi. Birds with lower loads of *S.* Enteritidis had increased activity in key immunological pathways associated with chemokine signaling, ErbB signaling, Jak-Stat signaling, and MAPK signaling compared with birds that maintained higher loads of *S.* Enteritidis colonization. In each of these signaling pathways, the birds with lower loads of *S.* Enteritidis had increased numbers of differentially phosphorylated peptides involved compared with the birds with higher loads of *Salmonella* Enteritidis. Further analysis identified BLNK, Raf1, and AKT3 as specific proteins that may be associated with increased resistance against *Salmonella* Enteritidis and could provide poultry breeders with additional biomarkers to identify birds naturally more resistant to this important foodborne pathogen potentially reducing the need for antibiotics and creating a safer food supply.

Key Words: *Salmonella*, kinome, peptide array, chicken, signaling

P110 Norepinephrine modulates swine gut immune cells.

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Norepinephrine (NE) is a hormone released in stressful conditions. This neuro-chemical is linked to gut microbiomes and immune modulation. NE is known to stimulate the growth of a wide spectrum of bacterial species, including *Salmonella* and *E. coli*, which can be present in the gastrointestinal tract of swine. Therefore, it is a risk for food safety. The objective of this study was to evaluate the effect of norepinephrine in the innate immune response in pigs. The data were analyzed by one way ANOVA with Scheffé's test. Mesenteric lymph nodes (MLN) were collected from healthy pigs and stimulated with norepinephrine for different lengths of time (10 min, 30 min, 1 h, and 2 h). The percentage of cells that ingested pathogens were increased by norepinephrine after 10 min of stimulation and remained at the maximum number when stimulated up to 2 h ($P < 0.01$). The associated oxidative burst, indicating killing activity, also increased by 10 min of stimulation ($P < 0.01$), but then gradually decreased with stimulation up to 2 h. There was no difference for cells positive for CD14 or CD18 with norepinephrine stimulation over the time points evaluated. However, CD18 had a tendency ($P < 0.10$) to increase linearly for 30 min then decreased to baseline with 1 and 2 h of stimulation. These data show a maximum effective time for stimulation of gut immune cells with norepinephrine. These preliminary data will be useful for the continued study of stress on gut immunity and demonstrate the quick response by MLN to norepinephrine. This could explain the fast shedding of *Salmonella* from pigs when subjected to stress conditions such as slaughter.

Key Words: norepinephrine, gut, swine, immunity, stress

P111 A role for the noncanonical Wnt-signaling pathway in the induction of a state of immune tolerance that allows the establishment of persistent intestinal colonization of *Salmonella enterica* serovar Enteritidis in chickens.

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Using a chicken-specific kinome peptide immune array, we have previously outlined the induction of a tolerogenic response in the cecum of chickens infected with *Salmonella* Enteritidis beginning around 4 d post-primary infection. The tolerogenic response is induced by a series of phosphorylation-mediated changes in the ceca of chickens during the development of a persistent *Salmonella* infection. The tolerance is characterized by alterations in immune cell signaling: dephosphorylation of phospholipase c-γ1 [PLCG1] and NF-κB; phosphorylation of NFAT, inactivation of AP-1 and mTOR signaling pathways, increased phosphorylation of AMP-activated protein kinase [AMPK], and blockage of IFN-γ protection through the disruption of the JAK-STAT signaling pathway (dephosphorylation of JAK2, JAK3, and STAT4). Further analysis of the signaling pathways altered during the establishment of this tolerogenic state has revealed a role for the noncanonical Wnt signaling pathway. The glycoprotein Wnt5 caused an influx of intracellular Ca^{2+} that led to the activation of Ca^{2+}-dependent effector molecules such as calcium/calmodulin-dependent kinase II (CamKII), calcinuerin, protein kinase C (PKC), JUN N-terminal kinase (JNK) and activates the phosphorylation of the transcription factor, NFAT. Nuclear translocation of NFAT resulted in a significant increase in the mRNA expression of the anti-inflammatory cytokines IL-10 and TGF-β. These studies describe further phenotypic changes from a pro-inflammatory environment during the acute phase of infection (4–48 h post-infection) to an anti-inflammatory, tolerogenic environment in (4 d post-infection) the avian cecum of *Salmonella* Enteritidis infected chickens that results in the establishment of persistent intestinal colonization.

Key Words: *Salmonella*, tolerance, intestine, anti-inflammatory

P112 Anti-interleukin-10 antibody is effective at eliminating the adverse effects of a coccidiosis challenge.

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Coccidia infection has been shown to increase intestinal interleukin-10 (IL-10) mRNA expression. Recent findings that IL-10 protein is released intraluminally and that there are apical receptors for IL-10 on the enterocyte suggests that coccidia evade the immune system by upregulating IL-10 secretion. IL-10 as a critical host molecule for protozoa pathogenicity is supported by reports showing that IL-10 knockout mice are more resistant to cryptosporidium infection than wildtype mice. We hypothesized that neutralization of intraluminal IL-10 would reduce the adverse effects of coccidiosis on chick performance without adverse effects on immunity to coccidia. To test this hypothesis egg antibody to chicken IL-10 peptide was fed as a dried egg yolk preparation (0.341 g/kg feed) to determine broiler growth response following a mild coccidia infection. Using a 2 × 2 factorial design, chicks were fed either anti-IL-10 or control antibody, then challenged at d 3 (d3) orally with either sterile saline or a 10× attenuated coccidia vaccine (Advent, Novus Intl.). Weights were measured 1 wk post vaccination as well as 18 d post vaccination. Coccidia infection in control antibody fed chicks reduced body weight 10% compared with non-infected control antibody fed chicks. Coccidia infected chicks fed anti-IL-10 had similar weight as control antibody non-infected chicks (Antibody × Infection interaction, $P < 0.05$). Oocyte shedding was also reduced in infected chicks fed anti-IL-10 ($P = 0.10$). In a second experiment, chicks were either fed anti-IL-10 or control antibody. Half the chicks in each group were either vaccinated at d 3 with a 1× dose of coccidia vaccine then challenged with a 10× dose of vaccine on d 17. The other half served as unvaccinated non-challenged controls (2 × 2 factorial with main effect of Antibody and Coccidia exposure). Coccidia exposure of control antibody fed chicks decreased body weight 6%, whereas chicks fed anti-IL-10 had body weights similar to those not exposed to coccidia (Antibody × Coccidia exposure interaction $P < 0.05$). These results suggest that neutralization of intraluminal IL-10 protects against the adverse effects of coccidosis.

Key Words: interleukin-10, coccidiosis, weight gain, oocyte, treatment

P113 Comparison of anti-interleukin-10 egg antibody to Maxiban in coccidia infected broiler chicks.

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Poultry producers lose and spend a total of one billion dollars a year due to coccidiosis and its control. Coccidosis is managed by either vaccination or the use of anti-coccidials (ionophores or chemical). Vaccines tend to reduce weight gain or offer limited protection against subsequent exposure, while the use of anti-coccidials creates both coccidia and public resistance. Recently we showed that anti-IL-10 was effective in reducing the adverse effects of coccidia challenge. In this experiment we compare the usefulness of the anti-coccidial, Maxiban to anti-IL-10. Chicks were fed a commercial diet with and without Maxiban, with and without anti-IL-10 (0.341 g/kg of feed as egg yolk powder), and with or without 10x dose of Advent coccidiosis vaccine (*Eimeria acervulina, Eimeria maxima*, and *Eimeria tenella*) at 3 d of age (2 × 2 × 2 factorial arrangement of treatments). Ten pens of 5 chicks were assigned to each of the 8 treatments in battery brooders with raised wire floor. Chick weights were taken on d 1, 3, 7, 10, 14, and 21, and the feed from each pen was weighed on d 1, 7, 14, and 21. Fecal samples were collected on d 11 to count *Eimeria* oocytes via the Modified McMaster's technique. At 2 and 3 weeks, a coccidiostat × antibody × challenge interaction was observed ($P < 0.05$). Chicks fed Maxiban and infected with coccidia had a reduced body weight (9% at wk 2 and 11% at wk 3). Reduced body weight due to coccidia infection in the Maxiban treated chicks was prevented when the diets included anti-IL-10. By wk 3, BW of coccidian-infected chicks fed Maxiban plus anti-IL-10 was 9% greater than chicks not infected and not fed either additive. Oocyte counts were decreased by coccidiostats ($P = 0.0017$) and anti-IL-10 ($P = 0.10$) in infected animals. No oocytes were found in control, PBS-gavaged, birds. In conclusion, feeding anti-IL-10 is equivalent to, if not better than, Maxiban in counteracting the adverse effects of coccidiosis on growth.

Key Words: coccidiosis, interleukin-10, *Eimeria*, broiler, Maxiban

P114 Effects of dietary fiber on cecal short-chain fatty acid and microbial community of broiler and layer chicks.

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An experiment was conducted to evaluate the effects of feeding various concentrations of dietary fiber on cecal short-chain fatty acid (SCFA) concentration and microbial communities of broiler and layer chicks. The lower fiber diet was based on corn-soybean meal (SBM) and the higher fiber diet was formulated using corn-SBM-dried distillers grains with solubles (DDGS) and wheat bran to contain 60.0 g/kg of both DDGS and wheat bran from 1 to 12 d and 80.0 g/kg of both DDGS and wheat bran from 13 to 21 d. Diets were formulated to meet or exceed NRC nutrient requirements for starter chicks. Broiler and layer chicks were assigned to battery cages within line to minimize differences in mean cage bodyweight. Treatment groups were randomly assigned to battery cages in a completely randomized design with 11 replicates of 8 chicks for each of the 4 treatments. Three ceca were collected from each replicate, one cecum underwent SCFA concentration analysis and the other 2 ceca underwent bacterial DNA isolation for terminal restriction fragment length polymorphism (TRFLP) and shotgun metagenomics analyses, respectively. The results indicated an interaction between bird line and dietary fiber for acetic acid ($P = 0.04$) and total SCFA ($P = 0.04$) concentration. There was a higher concentration of acetic acid ($P = 0.02$) and propionic acid ($P < 0.01$) in broiler chicks compared with layer chicks. Increasing dietary fiber resulted in a reduction of butyric acid ($P = 0.03$). Percent molar proportion of individual SCFA was not different between broiler and layer chick lines. Higher dietary fiber resulted in lower percent molar butyric acid concentration ($P = 0.02$). TRFLP analysis showed that cecal microbial communities varied due to diet ($P = 0.02$) and bird line ($P = 0.03$). Shotgun metagenomics analysis identified differences in the relative abundance of the species *Helicobacter pullorum* and

Megamonas hypermegale and of the genera *Enterobacteriaceae*, *Campylobacter*, *Fecalibacterium*, and *Bacteroides*. These results provided insights into the differences in effects of dietary fiber on SCFA concentration and modulation of cecal microbiota in broiler and layer chicks.

Key Words: broiler, layer, high-fiber diet, short-chain fatty acid, cecal microbiome, shotgun metagenomics

P115 Effects of *Saccharomyces cerevisiae* fermentation products on fiber digesting and lactate utilizing rumen bacteria at neutral and low pH in vitro.

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A meta-analysis of peer-reviewed publications reported that *Saccharomyces cerevisiae* fermentation products (SCFP) increase milk yield and dry matter intake in lactating dairy cows (Poppy et al., 2012). Rumen microbial population changes may help to explain these outcomes. Three in vitro experiments were conducted to evaluate the effects of various SCFP on concentrations of fiber digesting (FDB) and lactate utilizing bacteria (LUB) at neutral (6.7–6.9) and acidic (5.5–5.8) pH. Five samples of whole rumen contents obtained from a rumen-cannulated, nonlactating Jersey cow were composited, strained, buffered and dispensed into vessels containing a dried ground (1 mm) substrate (50:50, forage:concentrate) and treatment. Treatments were Control, XPC (Diamond V Original XPC), Prototype 1 (P1) and Prototype 2 (P2). Each experiment had 5 replicates/treatment. The vessels were sealed and incubated with continuous mixing at 39°C for 12 h. Acidic pH was achieved in experiments 2 and 3 by increasing the substrate 2.5 fold and diluting the buffer by 50%. The FDB (*Ruminococcus flavefaciens, Ruminococcus albus* and *Fibrobactersuccinogenes*), and LUB (*Megasphaera elsdenii* and *Selenomonas ruminantium*) were enumerated by real time PCR. Data are presented as fold changes relative to Control. At neutral pH (Exp. 1), XPC increased ($P < 0.05$) all FDB and *S. ruminantium*; larger fold changes ($P < 0.05$) were demonstrated by P2. P1 increased ($P < 0.05$) *R. flavefaciens* and *S. ruminantium*. *M. elsdenii* was not detected at neutral pH. At acidic pH (Exp. 2 and 3), XPC increased ($P < 0.05$) *R. flavefaciens* and *F. succinogenes* in both experiments, but decreased ($P < 0.05$) *S. ruminantium* in Exp. 3. P1 increased ($P < 0.05$) *R. flavefaciens* and *M. elsdenii*. P2 increased ($P < 0.05$) *R. flavefaciens*, *F. succinogenes*, and *M. elsdenii* in both experiments, but *S. ruminantium* in Exp. 2 only ($P < 0.05$). Results suggest that Original XPC enhances fiber digesting and lactate utilizing rumen bacteria at both neutral and acidic pH, and that these microbial responses are further magnified by Prototype 2.

Key Words: *Saccharomyces cerevisiae*, rumen bacteria, in vitro, PCR

P116 Synergistic effect of *Bacillus licheniformis* and Flavomycin on broiler performance.

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Direct-fed microbials (DFM) offer potential to ameloriate the intestinal imbalance following antibiotic use. However, in some instances; for example, high intestinal disease pressure, it might be preferable to run a DFM and antibiotics. This study was intended to demonstration DFM/antibiotic compatibility. A total of 1980 Cobb 500 male chicks were allocated to 5 treatment groups, 33 chicks per pen (12 replicates/treatment). All birds were fed a basal corn-soy based diet. Treatments were: 1) Non-infected Control (nCON); 2) Infected control (iCON; 3) *Bacillus licheniformis* (BL); 4) Flavomycin(FLV); 5) BL and FLV. FLV was administered at 2 ppm and BL at 1.6×10^6 cfu/g feed. At 7 d, old litter was introduced into each pen. At d 31, each bird in treatments 2–5 were orally administered 1 mL *Clostridium pefringens* (10^9 cfu/ml). Performance was recorded and NE lesion score determined at d 35. On d 35, all birds had NE lesions including nCON due to reused litter. BL+FLV had s lower NE lesion scores than all other treatments. FCR, liveweight and mortality did not differ significantly between treatments. However, BL and BL + FLV were numerically heavier at all time pointswhile D42 FCR was lowest in FLV and uCON. BL+FLV had the highest mortality although overall mortality did not differ significantly between treatments. Historically at the study site, a *C perfringens* oral dose of 10^6 cfu/mL in birds 2–3 weeks of age was adequate to demonstrate mild to moderate morbidity. This study used a *C. perfringens* 3 log higher in 5-wk-old birds, indicating that older birds have a higher degree of natural resistance to this strain of *C. perfringens*. This study demonstrates that BL can be used synergistically with FLV to improve bird performance greater than DFM or FLV alone under conditions of intestinal stress.

Table 1. Zootechnical performance

	Liveweight (lb)			FCR		Lesion	Mortality (%)	
	d 21	d 35	d 42	d 35	d 42	score	d 0-35	d 0-42
uCON	1.724	3.117	4.592	1.772	2.02	1.03^{ab}	1.93	2.48
iCON	1.711	3.049	4.545	1.781	2.06	1.39^{a}	3.20	3.58
BL	1.729	3.186	4.699	1.754	2.04	1.06^{ab}	2.66	3.25
FLV	1.725	3.116	4.462	1.795	2.02	1.03^{ab}	2.41	3.54
BL +FLV	1.741	3.217	4.691	1.750	2.03^{a}	0.83^{b}	4.37	4.67

Key Words: probiotic, poultry, bacillus, antibiotic, performance

P117 Effect of quercetin on performance, apparent digestibility of feed nutrients and cecal microbiota in laying hens at 39 to 47 weeks old.

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This trial was conducted to evaluate the effect of quercetin on performance in laying hens at 39–47 weeks old by determining laying rate, feed-egg ratio, apparent digestibility of feed nutrients and the amount of cecal microbiota. A total of 240 healthy Hessian laying hens at 39-week-old with similar body weight and laying rate were randomly divided into 4 groups with 6 replicates of 10 each replicate, respectively. The laying hens were fed with corn-soybean basal diet supplemented with 0, 0.02, 0.04, and 0.06 g quercetin /kg diet for 8 weeks. The results showed that compared with control, laying rate was significantly increased (by 7.14%; $P < 0.01$) and feed-egg ratio was significantly lowered (by 11.72%; $P < 0.01$) at 0.04 g/kg quercetin in laying hens at 39 to 47 weeks old. Apparent digestibility of crude protein and Ca were significantly increased (by 52.9% and 18.85%, respectively;

$P < 0.01$) at 0.04 g/kg quercetin in laying hens at 39 to 47 weeks old. The amount of total aerobic bacteria was significantly decreased ($P < 0.01$) by 0.04 g/kg quercetin in laying hens at 39 to 47 weeks old. The amount of *E. coli* was decreased and the amount of *Bifidobacterium* was significantly increased by 0.04 g/kg quercetin in laying hens at 39 to 47 weeks old. In conclusion, a certain dose of quercetin promoted *Bifidobacterium* growth and inhibited growth of total aerobic bacteria and *E. coli* in cecum of laying hens at 39 to 47 weeks old, increased availability of dietary crude protein and Ca, and therefore, improved performance of laying hens. The optimum level of quercetin to improve performance was 0.4 g/kg in the basal diet.

Key Words: quercetin, laying hen, performance, apparent digestibility, cecal microbiota

NOTES

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